STOP-Colitis Pilot Trial: A prospective, open-label, randomised pilot study to assess two possible routes of Faecal Microbiota Transplant (FMT) delivery in patients with ulcerative colitis.

STOP-COLITIS PILOT Trial Protocol

Version 4.0 17th August 2018

IMPORTANT NOTE

The attached STOP-Colitis published protocol relates solely to the pilot phase of the main STOP-Colitis RCT trial entitled:

'A double blinded randomised controlled trial to investigate the efficacy of faecal microbiota transplantation (FMT) in achieving and maintaining remission for patients with ulcerative colitis 13/179'

Once the STOP-Colitis pilot phase is complete, a separate protocol for the main RCT trial will be published, once all regulatory approvals are in place.

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EudraCT number:	**2015-005753-12**
Sponsor reference number:	**RG_15-026**
ISRCTN number:	**74072945**
IRAS ID number:	**170888**
REC reference number:	**17/EM/0274**
CPMS ID number:	**35522**

PROTOCOL DEVELOPMENT AND SIGN OFF

PROTOCOL CONTRIBUTORS
This protocol was written in a joint effort by the Trial Management Group of the STOP-Colitis Pilot Trial

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This protocol has been approved by:	
Trial Name:	**STOP-Colitis Pilot Trial**
Protocol Version Number:	4.0
Protocol Version Date:	17th August 2018
CI Name:	Professor Tariq Iqbal
Trial Role:	Chief Investigator
Signature and date:	___ _____ 17th/ Aug / 2018

Sponsor statement:

Where the University of Birmingham takes on the role of Sponsor for protocol development oversight, the signing of the IRAS form by the Sponsor will serve as confirmation of approval of this protocol.

PI SIGNATURE PAGE

The undersigned confirm that the following protocol has been agreed and accepted and that the Principal Investigator agrees to conduct the trial in compliance with the approved protocol.

I agree to ensure that the confidential information contained in this document will not be used for any other purpose other than the evaluation or conduct of the clinical investigation without the prior written consent of the Sponsor.

This protocol has been approved by:

Trial Name:	**STOP-Colitis Pilot Trial**
Protocol Version Number:	4.0
Protocol Version Date:	17th August 2018
PI Name:	
Name of Site:	
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TRIAL SUMMARY

TITLE

STOP-COLITIS PILOT: Prospective, open-label, randomised pilot study to assess two possible routes of Faecal Microbiota Transplant (FMT) delivery via the naso-gastric (NG) route or by direct delivery to the colon (COLON) in patients with ulcerative colitis.

Trial Design

Prospective, open-label, multi-centre, randomised pilot study.

Objectives

The primary objective of the study is to assess the effectiveness and acceptability of the two routes (NG or COLON) of FMT delivery, and to determine whether FMT by either NG or COLON is suitable to take forward to a double-blinded randomised controlled trial (RCT) to investigate the efficacy of FMT (vs. placebo) in achieving and maintaining remission in patients with active ulcerative colitis.

Participant Population and Sample Size

The study will recruit a total of 30 patients (from age 16) with active ulcerative colitis.

Outcome Measures

The primary outcome from the pilot will be a recommendation about a route of administration to take forward to the main RCT. This decision will be based on comparing the two methods NG and COLON using a composite assessment of both quantitative and qualitative data. This will include assessment of efficacy (using clinical response), acceptability and safety.

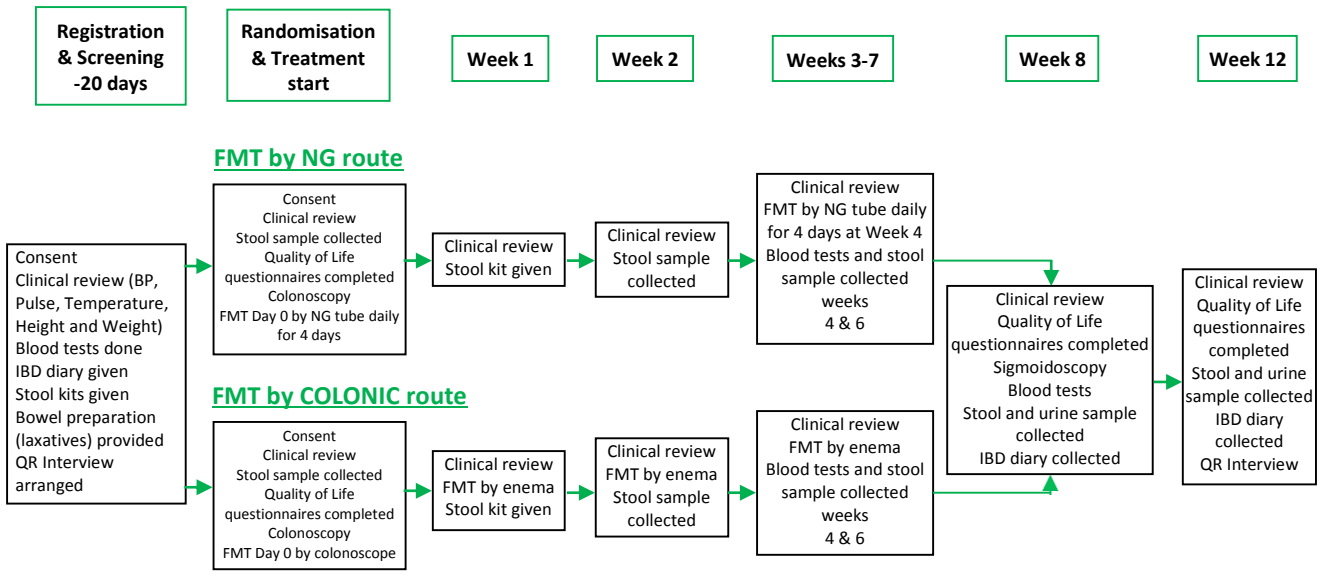
Key Eligibility Criteria

The trial will include patients (aged 16 or over) with ulcerative colitis diagnosed for at least 12 weeks. .They will have active disease as defined by a partial Mayo score of ≥4 and ≤8 despite stable maintenance medical treatment or no treatment.

Intervention

Patients will be randomised to receive FMT by either the NG or COLONIC route.

Trial Schema



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1. BACKGROUND AND RATIONALE

1.1. Background

The genetic material in the human microbiome contains 100 times more DNA than that found in human cells and the gut microbiome is essential for the maintenance of health. In the inflammatory bowel diseases (IBD), Crohn's disease (CD) and ulcerative colitis (UC), there is 'dysbiosis' of the gut microbiota compared to the healthy bowel, with IBD showing reduced bacterial diversity compared to that seen in healthy individuals [1,2]. In fact, the currently accepted paradigm concerning the pathogenesis of IBD involves an aberrant immunological response to intestinal bacteria in a genetically susceptible host. Whether the observed dysbiosis is the initiating abnormality or whether this is a secondary consequence of inflammation remains unanswered. Faecal Microbiota Transplant (FMT) is the infusion of a faecal suspension from a healthy donor into the gastrointestinal tract of a patient with disease, and there is great interest in the potential for modifying the gut microbiome as a potential treatment for IBD [3].

Since the first descriptions of CD and UC at the beginning of the twentieth century, it has been strongly suspected that the gut microbiota may have a defining role in the pathogenesis of IBD [4]. Early culture-based studies underestimated the complexity of the microbiota, and were hampered by various culture biases and the challenge of enumerating fastidious bacteria with challenging growth conditions. With the advent of cheap high-throughput genetic sequencing techniques allied with complex bioinformatics capability, there has recently been a revolution in our understanding of the colonic microbiome. As a result of studies on the microbiome both in patients with IBD and animal models, we now know that patients with IBD (either CD or UC) have reduced bacterial diversity compared to healthy individuals, and at the phylum level a reduction in Firmicutes and a relative increase in Proteobacteria [2,5]. The environmental contribution to the aetiopathogeneisis of UC is greater than that in CD. Data has suggested that alteration in the gut microbiome plays a central role in driving UC; datasets highlighting the importance of *Roseburia.hominis* [6], *Faecalibacterium.prausnitzii* [7] and *Akkermansia.muciniphila* [8] in the inflammation in UC have been published. Attempts to alter the microbiome with probiotics, whilst disappointing in CD, have shown promise in UC [9,10]. Although the data are limited to case series, the evidence for a positive efficacy signal for FMT is greater in UC than CD, with ten positive case series published [11]. There is a suggestion from both humans and animal models that the inflammatory effects associated with the dysbiosis may be due to differential carbohydrate and protein fermentation [12], and the metabolic end products of fermentation may modulate colonic epithelial inflammatory pathways [13]. The majority of the genes linked with IBD in recent genome wide association searches implicate bowel wall proteins, which act to signal between the colonic lumen and inflammatory pathways in the lamina propria [14]. Similarly, we have shown that major bacterial fermentation metabolites changed dramatically during induction of remission and reduction of colonic inflammation with exclusive enteral nutrition treatment [15,16]. Although the observed changes were against our *a priori* hypothesis, these data suggest that bacterial metabolic activity may be implicated in the aetiology of IBD and should be explored in future research with novel metabolomic technologies.

These advances in our understanding of the pathophysiology of IBD and the central role of colonic bacteria makes the manipulation of the colonic microbiome a potentially highly fruitful endeavour in the treatment of IBD. Early efforts to affect the colonic microbiota using probiotics have largely been disappointing, but these have been limited by products lacking bacterial diversity and concentration [9]. FMT, by contrast, involves the transfer of potentially an entire dense colonic microbial community. Data concerning the use of FMT in IBD are limited, but the signals are encouraging. Reports of FMT in IBD are generally individual case reports or small case series, and this technique has not yet been subject to rigorous clinical trials. In 2012, a meta-analysis reported that, from the small numbers studied, FMT achieved remission in over 50%. Encouragingly about three quarters of the patients were able, in the short term, to stop all other IBD related medication [3]. The majority of studies with FMT in IBD have been undertaken by delivering the transplant directly into the colon either through the colonoscope or using enemas. Delivery of the faecal transplant into the upper gut via a naso-gastric tube into the stomach is an alternative and *Clostridium difficile* treatment studies have shown that use of either route seems equally effective [17]. Naso-gastric (NG) FMT has been used in two small cases series of patients with UC and CD. In the former, the response was much less (20%), and there were some self-limiting systemic side effects [18]. However, these patients were all at the most severe end of the spectrum of UC having failed "rescue" therapy with powerful biologic agents. Furthermore, these patients all stopped immunosuppressive medication

before FMT. The one patient who did respond showed a very good response with a reduction in Mayo score from 11 to 3 at three months after the procedure. Furthermore, the authors were able to correlate clinical response to reduction in Enterobacteriaceae [18]. It may be argued that these patients were beyond medical intervention and perhaps unlikely to respond to any treatment at that stage. However, in spite of this, a positive treatment signal was seen. With regard to the question of stopping immunosuppressive treatment before using FMT, a retrospective case series published in 2014, provides reassuring data for the safety of FMT in immunocompromised patients [19]. In four patients with CD, a single 200g dose of FMT was administered after colonoscopy via an NG tube. Again these patients were at the severe end of the spectrum of disease, the signal (change in microbiome) was transitory and there were self-limiting side effects [20]. The effect of FMT in CD may be complicated by the small intestinal ulceration associated with this type of IBD which may allow translocation of bacteria outside the gut and makes FMT less attractive and potentially more hazardous than is the case with UC. There has been only one randomised controlled trial (RCT) using rectal enema FMT compared to placebo in the treatment of UC [21]. This was presented at the Digestive Diseases Week in Chicago (2014). Interim results showed a 23% remission rate compared to placebo (7%), and despite highly encouraging signals in individual patients, the trial was stopped early by the Independent Oversight Committee as it was deemed unlikely to reach its primary endpoint. This study had a highly ambitious primary endpoint of complete mucosal healing which was only achieved in four (15%) patients in the transplant arm and two (8%) in the placebo arm. Another potential flaw was the utilisation of a somewhat "limiting" treatment regimen of 6x weekly enemas with no colonoscopic delivery, despite the fact that the majority of study entrants had pancolitis. This would greatly limit the possibility of modulating the microbiota, particularly on the right side of the colon. Despite the "negative" clinical data, there were significant microbial and metabolomic changes in the clinical responders [22].

We currently have a clinical protocol which we use for treating patients with recurrent *Clostridium difficile* infection (CDI) related colitis with FMT via the NG route. We intend to use this approved methodology to supply FMT and adapt it to study the two possible routes of FMT delivery. This becomes feasible following data that demonstrates that freeze-thawing stool does not lead to deterioration in the microbiome in the time frame mandated for the current study. This is evidenced by two studies which indicate that frozen stool kept for three months is effective at treating CDI [17,23], and it has now been shown that freezing donated stool for up to six months does neither result in a significant deterioration in bacterial diversity nor reduction in the efficacy in treating CDI [24]. In the former, the investigators showed that despite freezing, the bacterial diversity following treatment resembled that of the donor before the sample was frozen [20].

Data regarding FMT as a treatment for UC are limited at this time and numerous questions regarding treatment protocol remain to be answered; for example what is the optimum method of donor/ patient selection and preparation and the best route of delivery? In addition to the variation in route of administration, the dose of faecal transplant administered has varied greatly among the small patient series treated to date. Generally speaking, the best treatment signals are seen in those studies where intensive regimens have been used. In the current trial, we propose an intensive treatment regimen with patients receiving 240g by the NG and 360g via the colonic route over the treatment period. This is more than in previous studies in patients with IBD.

This protocol describes the first part (the pilot study) of a two-stage study:

1. Pilot study to investigate the optimum route of FMT delivery for the treatment of UC. This pilot study will assess the effectiveness and acceptability of the two routes of FMT delivery (the naso-gastric route of delivery and delivery directly to the inflamed colon), and determine whether FMT by either NG or COLON is suitable to take forward to a main RCT. We would then plan to take one of these protocols (given a suitable treatment effect) forward to a main RCT powered to assess the efficacy of this treatment.
2. Double-blind placebo-controlled RCT to investigate the efficacy of FMT in achieving and maintaining remission for patients with UC.

1.2. Trial Rationale

1.2.1. Risks and benefits

The main theoretical risk associated with FMT is the transmission of exogenous infections to patients many of whom are already immune-compromised in relation to their IBD treatment. To minimise this risk, we will use the careful donor risk assessment and screening regimen currently in place in our Human Tissue Act (HTA)-approved CDI treatment programme, which conforms to American Gastroenterology Association (AGA) guidelines [25] and will be licenced by the UK competent authority, the Medicines and Healthcare products Regulatory Agency (MHRA), for this trial. This ensures robust donor suitability and traceability of the donor stool. As of January 2017, within this programme, there have been no cases of septicaemia linked to FMT in over 150 cases treated. In addition, as part of the treatment protocol, we will carefully analyse donor and recipient stool and colonic mucosa for microbiome and metabolomic changes associated with clinical outcomes. In order to minimise the potential for septicaemia, we have elected to confine this study to UC and not include patients with CD, who have deeper, transmural ulceration and are probably more prone to extra-luminal sepsis/ bacterial translocation across the gut. Early reports also suggest that patients with CD are perhaps more likely to suffer from side effects with FMT [20]. To minimise the risk of regurgitation and aspiration via the NG route, we will give FMT after an overnight fast, co-administering a pro-kinetic and a proton pump inhibitor to encourage gastric transit and reduce the volume of gastric secretions respectively.

1.2.2. Justification for participant population

UC is a chronic debilitating disease with an increased risk of bowel cancer and management relies on life-long anti-inflammatory/immunosuppressive agents and, in some cases, patients need colectomy. UC generally affects young people who are in the reproductive and working stage of life. Thus, quite apart from the costs associated with investigations, long-term follow-up, hospital admissions, toxic anti-inflammatory drugs and surgery, chronic UC is associated with a significant societal cost relating to loss of earnings and productivity in relation to disease exacerbations.

Patients with UC who are in remission on medical treatment have a 50% chance of a flare of their disease every year, and a third of patients will eventually need colectomy [26]. There is a pressing need for better and more effective medical treatments to reduce the need for surgery and reduce the long-term cancer risk. In this regard, FMT represents a very low-cost, potentially paradigm-changing, treatment for UC. In theory this treatment would, for the first time, address the cause of the disease rather that reactively treating the resulting inflammation. This, therefore, presents a potentially curative approach.

1.2.3. Justification for design

This protocol describes the first part of a planned 2-stage investigation to study firstly the optimum route of FMT delivery for the treatment of UC, and secondly (subject of a separate protocol), the efficacy of such treatment in patients with active UC which is refractory to standard treatment. The initial pilot study described here will assess both NG delivery of FMT and delivery of FMT directly to the colon (COLON). At the end of this pilot, there will be a STOP/GO point at which time an independent oversight committee will decide (1) which (if any) of the two routes of FMT delivery to take forward to the main trial; and (2) whether a full RCT is feasible. These decisions will be based on a pragmatic review of the pilot data in terms of assessing treatment efficacy (based on clinical response), tolerability, patient adherence to allocated treatment and qualitative perspectives on FMT and trial experience. Stop-Go guidelines will be used to determine whether to proceed forward to the main RCT, and, we propose then to take forward the preferred method into a randomised double-blind, placebo-controlled trial with an efficacy outcome measure of clinical remission.

1.2.4. Choice of treatment

We had the first HTA-approved treatment for *Clostridium difficile* related colitis and, as such, a well-developed protocol for donor screening and transplant preparation. The newly set-up and dedicated University of Birmingham Microbiome Therapy Centre (UoBMTC) will be using FMT produced under an MHRA licence to study the two possible routes of FMT delivery (NG or COLON).

1.2.5. Sub-studies

This study will afford us the possibility of embedding scientific sub-investigations which will provide considerable information regarding alterations in the microbiome and associated metabolic pathways which may be linked with clinical efficacy. We will also be able to look at characteristics of donor stool, donor dietary habits and associate these with clinical effects.

2. AIMS, OBJECTIVES AND OUTCOME MEASURES

2.1. Aims and Objectives

The aims of this pilot study are:

i) To determine which FMT administration route (NG or COLON) should be investigated in a double-blind, placebo-controlled RCT; and

ii) To determine if a full RCT is feasible.

2.1.1. Clinical Objectives

To assess:

- Whether FMT by the NG route induces clinical response in patients with active UC;
- Whether FMT by the colonic route induces clinical response in patients with active UC;
- Tolerability and safety;
- Which route of FMT delivery (if any) is suitable to investigate in the RCT.

2.1.2. Qualitative Objectives

To assess:

- Patient acceptability of FMT (NG);
- Patient acceptability of FMT (COLON).

2.1.3. Mechanistic Objectives

To assess:

- Whether FMT by either route is associated with a change in faecal calprotectin as a surrogate marker of colonic inflammation;
- Changes in the colonic microbiome and metabolome (short chain fatty acids (SCFA)) induced by FMT via each route;
- Reduction in C-reactive protein (CRP);
- Engraftment of donor microbiota in recipients by culturing donor stool to strain level

2.1.4. Other Objectives

- Effect of diet (donors);
- Time from stool donation to treatment.

2.2. Outcome Measures

The primary outcome will be a composite assessment of both qualitative and quantitative data based on efficacy, acceptability and safety.

2.2.1. Clinical Outcome Measures

- Clinical response (primary measure of efficacy) defined as ≥3 point reduction in the full Mayo score from randomisation to week 8, and 30% reduction from randomisation and at least 1 point reduction of rectal bleeding subscore or an absolute rectal bleeding subscore of 0 or 1;
- Time to clinical response (where clinical response is defined as ≥2 point reduction in partial Mayo);

- Clinical remission at week 8 (full Mayo score of ≤2, with no subscore >1);
- Participant's weight at week 8 and week 12;
- Quality of Life (QoL) using generic Short-Form 36 (SF-36) and the disease specific Inflammatory Bowel Disease Questionnaire (IBDQ) at week 8 and week 12;
- Adherence to FMT;
- Adverse events (AE) and serious adverse events (SAE).

2.2.2. Qualitative Outcome Measures

Patient acceptability will be assessed through qualitative research interviews, see section 8.3.

2.2.3. Mechanistic Outcome Measures

- Faecal calprotectin;
- Measures of microbiome (faecal and mucosal);
- Mucosal healing;
- Urinary metabolome (SCFA);
- CRP.
- Donor faecal culture

2.2.4. Other Outcome Measures

- Association between the donor's dietary profile and microbiome;
- Time, i.e. number of days from donor stool processing to treatment of the patient (recipient) at Site for association between efficacy and freezer life of FMT.

3. TRIAL DESIGN AND SETTING

3.1. Trial Design

The **STOP-Colitis** pilot study is a multi-centre, open-label RCT of FMT delivered by the NG route versus direct delivery to the COLON in 30 patients with active UC.

At the end of the pilot study, data will be reviewed by an Independent Oversight Committee (IOC) who, according to pre-specified STOP/GO criteria (see section 13.5), will decide (1) which route of FMT administration (NG or COLON) is most appropriate to investigate in the double-blind, placebo-controlled RCT and (2) whether it is feasible to proceed to the main RCT.

3.2. Trial Setting

3.2.1. Clinical setting

Participants will be recruited from hospitals in two Local Clinical Research Networks (LCRNs) in England and a hospital in the Greater Glasgow area. Potential study participants will be identified when presenting for their routine hospital clinic visits. Members of site staff will screen for potential eligible study participants using the inclusion/exclusion criteria. Patients who fulfil the inclusion criteria will have their eligibility confirmed by medically qualified personnel with access to and full understanding of their medical history. After confirming eligibility, eligible patients will be approached by an appropriately trained member of the clinical team to ascertain interest in entering the study. If the patient wishes to participate, they will be invited to attend hospital during which written informed consent will be obtained from the patient by medically qualified personnel. Consent may also be taken at the study screening visit. All study procedures from the screening visit onwards will be carried out at the clinical research facilities at Birmingham, Glasgow and London respectively.

3.2.2. Donor setting

Donors will be recruited by advert from healthy individuals in Birmingham but we exclude healthcare workers due to their potential exposure to microbes affecting the microbiome. All donor related procedures including screening questionnaires, blood and stool collections will be carried out at the National Institute for Health Research (NIHR) Wellcome Trust Clinical Research Facility (WTCRF) at the Queen Elizabeth Hospital, Birmingham. Collected donations will be transferred to the UoB, where the donor stool will be prepared according to our standardised protocol and will be delivered to the three sites carrying out the pilot (Birmingham, Glasgow and London).

4. ELIGIBILITY

4.1. Inclusion Criteria

1. Clinically confirmed UC for at least 12 weeks prior to the screening visit;
2. Aged 16-70 years;
3. Partial Mayo score of ≥4 and ≤8 despite stable 5ASA+/- thiopurine, methotrexate or no treatment;
4. Rectal bleeding subscore of ≥1 on the partial Mayo;
5. Written, signed informed consent to the study.

4.2. Exclusion Criteria

1. Stool positive for *Clostridium difficile* or infection by either PCR or ELISA;
2. Positive for Hepatitis A/B/C, and/or Human Immunodeficiency Virus (HIV) infection;
3. Antibiotics in the preceding 12 weeks prior to date of the screening visit;
4. Systemic/topical steroids in the preceding 2 weeks prior to the date of the screening visit;
5. Biologics in the preceding 12 weeks prior to the date of screening visit;
6. Commercial probiotics and prebiotics in the preceding 12 weeks prior to the date of the screening visit;
7. On oral nutritional supplements or enteral/parenteral nutrition in the preceding 4 weeks prior to the date of the screening visit;
8. Pregnant or lactating. Note: *Spot urine will be performed at screening and randomisation to rule out pregnancy in females;*
9. Not willing to take appropriate contraceptive measures to prevent pregnancy during trial participation:

- Female participants of child bearing potential, unless willing to use two forms of contraception, one of which must be a barrier contraception (e.g. female condom or occlusive cap (diaphragm or cervical vault/caps) with spermicide) during the study and for 30 days from the date of last FMT dose (however a male condom should not be used in conjunction with the female condom);
- Male participants whose partner is of child bearing potential, unless willing to use an appropriate barrier method of contraception (condom and spermicide) in addition to having their female partner use another form of barrier contraception (e.g. occlusive cap (diaphragm or cervical vault/caps) with spermicide) during the study and for 30 days from date of last FMT dose (however a male condom should not be used in conjunction with a female condom).

5. CONSENT

5.1. Trial Participant Consent

It will be the responsibility of the Local Principal Investigator (PI) to ensure that written informed consent is obtained for each participant prior to performing any trial related procedures. Consent must be taken either by a medically qualified doctor, i.e. Investigators to include the Local PI, the clinical research fellow appointed to help run the study or co-investigators (Good Clinical Practice (GCP) trained Consultant Gastroenterologists,

Research Fellows) as delegated by the local PI and captured on the Site Signature and Delegation Log. Research nurses, if delegated as per the Site Signature and Delegation Log, may be involved in the informed consent process by way of introducing and discussing the study to patients, but it will be the responsibility of the PI to ensure that written informed consent is undertaken by a medically qualified doctor.

A Participant Information Sheet (PIS) will be provided to facilitate the consent process. Investigators will ensure that they adequately explain the aims and purpose of the pilot study, trial treatment, anticipated benefits and potential hazards of taking part in the trial to the participant. They will also stress that participation is voluntary and that the participant is free to refuse to take part and may withdraw from the trial at any time. The participant will be given adequate time to read the PIS, and to discuss their participation with others outside of the site research team allowing them ample time to consider the trial. The participant will be given the opportunity to ask questions and will be provided with the contact details of their local IBD research team should they wish to discuss the trial after the hospital visit.

If the participant expresses an interest in participating in the trial, they will be consented via a two-stage consent process as described below.

Stage 1. Consent for screening

The first stage will involve consent to the study for trial-specific screening activities and for consent to collect clinical samples for the mechanistic sub-studies. The patient will be invited to attend a hospital screening visit and asked to sign and date the latest version of the Screening Informed Consent Form (SICF). The Investigator will then sign and date the relevant consent forms at the same consent appointment. A copy of the SICF will be given to the participant, a copy will be filed in the medical notes, a copy sent to the **STOP-Colitis** Trial Office and the original placed in the Investigator Site File (ISF). Once the participant is registered into the trial (via telephone call to the **STOP-Colitis** Trial Office), the participant's details will be forwarded by the **STOP-Colitis** office to the qualitative researcher to enable the first qualitative interview to be conducted before randomisation and thus prior to the first FMT treatment visit.

If any patients decline to participate in the study, they will be offered the opportunity to take part in an interview with the qualitative researcher. Patients will be asked for consent for the qualitative interview (Section 8.3).

Details of the informed consent discussions will be recorded in the participant's medical notes in accordance to GCP. This will include date of discussion, the name of the trial, summary of discussion, version number of the PIS and SICF given to the participant. As consent will be obtained on the same day that the trial-specific screening activities are due to start, i.e. the screening visit, a note will be made in the medical notes to clearly indicate that consent was obtained prior to undertaking any study assessments.

Stage 2. Consent for trial entry

The second stage of consent is for entry into the trial.

The patient must be consented for trial entry and randomisation on the same day; this will occur when the patient attends the IBD clinic after taking the bowel preparation (see Section 8.0 Trial Procedures). Consent will be taken by the Investigator using the latest version of the Informed Consent Forms (ICF). The Investigator and patient will sign and date the relevant consent form at this visit. A copy of the ICF will be given to the participant, a copy will be filed in the medical notes, a copy sent to the **STOP-Colitis** Trial Office and the original placed in the ISF. Once the participant is randomised into the trial, the participant's unique trial number will be entered on the ICF and this will be maintained in the ISF.

All details of the informed consent discussions will again be recorded in the participant's medical notes in accordance to GCP as described under consent taken for screening.

At each visit, the participant's willingness to continue in the trial will be ascertained and documented in the medical notes. Throughout the trial, the participant will have the opportunity to ask questions about the trial. Any new information that may be relevant to the participant's continued participation will be provided. Where new information becomes available which may affect the participants' decision to continue, participants will be given time to consider and if happy to continue they will be re-consented. Re-consent will be documented in the medical notes. The participant's right to withdraw from the trial will remain.

Electronic copies of the PIS and ICF will be available from the **STOP-Colitis** Trial Office and will be printed on the headed paper of the local institution. Details of all participants approached about the trial will be recorded on the **STOP-Colitis** Participant Screening and Enrolment Log and with the participant's prior consent. The patient's General Practitioner (GP) will also be informed that they are taking part in the trial via a GP Notification Letter which will be sent to the patients GP from the randomising investigator.

5.2. Stool Donor Consent

FMT donor screening and donation is a well-established process in Birmingham for the clinical treatment of *Clostridium difficile* related colitis. However, as the donations in this instance will be in respect to this clinical trial, consent will be sought from the healthy volunteers prior to the donation of faecal material; a separate Donor Information Sheet and Consent Form is provided to inform donors of the trial. Consent for donors will be taken at the WTCRF by a **STOP-Colitis** Investigator (a medically qualified doctor) prior to the collection of any blood and stool samples taken from the donor for screening.

6. REGISTRATION AND RANDOMISATION

6.1. Enrolment

Participants will be recruited from hospitals in two LCRNs in England and a hospital in the Greater Glasgow area. The pilot study will take place at sites in Birmingham, Glasgow and London.

Currently, patients with UC under the care of gastroenterologists are seen regularly in a hospital out-patient clinic. Clinical research nurses and delegated investigators will screen for potential eligible study participants using the inclusion/exclusion criteria. Potential study participants will be identified by their IBD clinical team when presenting for their routine hospital clinic visits, reflecting the secondary care basis of the proposed research. Patients may also be identified from IBD multi-disciplinary team meetings, from local IBD databases or, occasionally, from patients in hospital with IBD. Patients who fulfil the inclusion criteria will have their eligibility confirmed by a medically qualified doctor with access to and full understanding of their medical history. After confirming eligibility, eligible patients will be approached by GCP trained clinicians (doctors and nurses) to ascertain interest in entering the study. This individual will give a comprehensive verbal explanation of the study (explaining both the two potential investigational and standard treatment options and highlighting any possible benefits or risks relating to participation). Time for questions throughout the discussion will be given and questions adequately addressed.

Participants who agree to enter the study will be asked to consent for entry into the study via completion of a Screening/Informed Consent Form for each stage of consent, i.e. screening and at randomisation.

Throughout the study, participants will be encouraged to ask questions and will be reminded that they can withdraw at any time without their clinical care being affected.

Recruitment is planned for a 3 month period followed by a 3 month follow-up period. This equates to around one patient per week from each site. To conform to CONSORT guidelines, participating centres will be requested to complete an anonymised log for all patients screened for eligibility, but who are not registered due to

ineligibility or because they decline further participation to randomisation. Reasons for declining will be sought as this will potentially inform recruitment to the subsequent RCT.

6.2. Registration

Following consent being taken, patients will be registered to **STOP-Colitis**. During registration, patient details and hospital details will be collected together with the name of the medically qualified doctor who obtained the written informed consent, the date the consent was taken and the versions of the PIS and SICF used to take consent. The patient will be issued a unique registration identification Number (RNO) which will be used for all correspondence relating to the patient.

6.3. Randomisation

Following confirmation of the patients *Clostridium difficile* result and the qualitative interview having taken place (see Section 8.0 Trial Procedures), patients will be invited to attend for randomisation and administration of the first FMT treatment. Randomisation to the **STOP-Colitis** treatment route will occur on the morning of this visit, i.e. when the patient attends the Clinical Research Facility following consent to randomisation and confirmation of all eligibility criteria by the Investigator on the day of randomisation. During randomisation, patient details and hospital details as collected at registration will be re-checked and confirmation that the patient meets the full eligibility criteria will be obtained.

Patients will be randomised at the level of the individual in a 1:1 ratio to either NG or COLON delivery of FMT. Randomisation will be provided by a computer-generated program at the Birmingham Clinical Trials Unit (BCTU). A minimisation algorithm will be used to ensure balance in the treatment allocation over the following variables:

- Partial Mayo score (4-5 or 6-8);
- Current smoking status (current smoker: yes or no†);

†Not smoked for the past 12 months.

A 'random element' will be included in the minimisation algorithm, so that each patient has a probability (unspecified here) of being randomised to the opposite treatment that they would have otherwise received. Full details of the randomisation specification will be stored in a confidential document at BCTU.

After randomisation, the patient's RNO (issued at Registration) will be referred to as the TNO, i.e. the trial identification number which will continue to be used for all correspondence relating to the patient.

7. TRIAL TREATMENT

7.1. Treatment

Within the **STOP-Colitis** pilot study, the treatment being given is Faecal Microbiota Transplant (FMT). For the purposes of the trial, FMT is regarded as an Investigational Medicinal Product (IMP).

7.2. FMT Supply and Storage

7.2.1. Donor selection and screening

FMT material will be prepared from stools provided by un-related, anonymous healthy donors. Donor inclusion/exclusion criteria will follow the AGA [25] recommendations. For inclusion, donors must be ≥18 and <50 years of age, have a normal morning bowel habit, have a normal body mass index (BMI≥18.5 and ≤25), be non-smokers (not smoking for at least 12 months) and have no recent history of diarrhoea or rectal bleeding. The full screening criterion is detailed in the **STOP-Colitis** Donor Screening Protocol (copy available via **STOP-**

Colitis Trial Office). Donors will be screened for blood-borne and enteric pathogens immediately prior to donation. Donations will then be prepared and frozen, and released for use subject to satisfactory screening results.

As **STOP-Colitis** involves an intensive treatment regimen, the current donor pool will need to be augmented considerably. The donor screening program in Birmingham will be extended to include staff members of the UoB and other members of the public who are not involved in healthcare, as donors on a re-imbursement basis. Stool will be pro-actively collected, frozen and stored, so as to ensure an adequate supply prior to the commencement of the trial. This prospective collection will continue throughout the trial, being mindful that the "freezer life" of the donor stool is 24 weeks [24]. For the purposes of this current pilot study, it will be necessary to stagger screening and recruitment of donors over the trial recruitment period given the limited "shelf life" of FMT. It is anticipated that at least 50 donors will be screened in order to recruit the 30 needed for the pilot trial.

7.2.2. Donor stool, stool collection, preparation and storage

Stool will be collected in sterile honey jars prospectively from donors over a time period of approximately ten days following donor screening at the WTCRF. Each fresh stool donation will be transported to the UoB for preparation and freezing at -80°C on the day of production. Donations not reaching the UoB within 6 hours of donation will be discarded and not used to prepare FMT. The FMT will be prepared at the dedicated containment level 2 laboratory at the UoBMTC within a Class 2 microbiology safety cabinet. All equipment used for FMT preparation will be dedicated for sole use for this purpose. Consumables used for preparation will be single use and sterile, and will be recorded on batch processing records, which will be stored at UoBMTC. Preparation of the donor stools will involve homogenisation and filtering in normal sterile saline, containing 10% glycerol, using sterile filter bags and mechanical homogenisation in a stomacher. Donor stool will be frozen with a glycerol cryoprotectant as published pilot data confirm microbial diversity and viability for up to 24 weeks at -80°C [24]. FMT material will be prepared in 50ml ready-to-use aliquots containing 30g of stool.

Stool filtrates will be stored ready for use at -80°C at UoBMTC, and will be given 24 weeks expiry from the date of production. An aliquot of all stool donations will be stored at -80°C indefinitely at UoBMTC for clinical governance and look back exercises. We will carefully record time from donation to treatment for all patients receiving FMT within this study. Each FMT treatment set prepared will be allocated a unique batch and lot number and cross-referenced to FMT preparation records and donor screening records. This will ensure full traceability of FMT material while on site at UoB. Batch records will be securely held at UoB.

FMT packaging and labelling

All FMT treatment material will be packaged and labelled before -80℃ storage on site at UoBMTC by GMP (Good Clinical Practice) trained qualified research scientists. FMT will be packaged in sterile 60ml (or similar) leak-proof containers, which are suitable for storage at -80℃. Individual containers will be further sealed with Parafilm M®, a plastic paraffin film, around the lid and packaged with absorbent material. Each FMT treatment pot will be labelled with a batch number, lot number, and expiry date and dispatched as a treatment set of 12x50ml FMT aliquots per patient.

Each FMT treatment set will be transported with a copy of the validation certificate, which will need to be retained in the participant's notes, for traceability and governance purposes. The validation certificate will include reference to all unique FMT identifiers (as described above) for traceability, will provide storage and shelf life instructions and will confirm all donor testing results. A copy of the QP batch release certificate will also be provided which will be signed by the Qualified Person (QP) prior to technical release of each treatment set to confirm that the treatment has met the batch release criteria for use of the FMT in the study. The FMT batch release criteria is described in the **STOP-Colitis** Donor Screening Protocol which is available from the **STOP-Colitis** Trial Office.

7.2.3. FMT (IMP) storage and supply

FMT storage at UoB

Prior to distribution to clinical sites, FMT will be stored in a -80℃ dedicated lockable freezer, located within an access controlled containment level 2 laboratory at UoBMTC. This freezer will be for the sole use of FMT material and will be monitored for temperature control as part of the existing quality management system at UoB. This includes the use of Tutela temperature monitoring system, which automatically records the freezer temperature every minute and alerts the laboratory to any deviation outside pre-set temperature limits. There is also -80℃ freezer capacity at UoB to ensure that if there is a freezer failure, contingency -80℃ freezer space would be available. There will be clear physical separation of FMT material in quarantine and material which has been released for use.

FMT supply and transport to recruiting sites

Prior to the start of recruitment, all sites will receive an initial supply of two treatment sets of 12 x 50ml aliquots FMT containing 30g of stool in each aliquot.

FMT material will be transported overnight by specialist courier (Biocare) on dry ice in secure, tamper evident containers under shipping code UN3373. Each container will be temperature monitored during shipment to ensure the product remains frozen during transport.

On receipt, individual sites will need to follow the FMT (IMP) Management Guidelines for FMT receipt, tracking and handling. The IMP Management Guidelines will be sent to Sites as part of the **STOP-Colitis** ISF.

FMT storage at recruiting Sites

When received at the clinical site, FMT material will be logged in the **STOP-Colitis** FMT Receipt and Treatment Accountability Log and transferred immediately to -80℃ storage in a dedicated lockable freezer, located within the local Clinical Trials Facility. As required for participant treatment, initially 5x50ml FMT aliquots will be removed from -80℃ storage, cross-referenced against the FMT Receipt and Accountability Log for treatment set number, batch number, lot number and expiry date, and then thawed at room temperature for 3 hours. Use of the FMT aliquot will be recorded in the FMT Receipt and Accountability Log and details also recorded on the validation certificate issued per FMT treatment set. Once thawed, FMT material will be transferred to the delivery receptacle (enteral feeding syringe for NG route, 5x50ml syringes for flushing into the colonoscope or enema bottle for COLON route) and taken immediately to the patient for administration. In the case of randomisation to the NG treatment, it is acknowledged that there will be some wastage for the first treatment due to logistical constraints with respect to time required to thaw FMT. Sites will need to document the number of aliquots discarded on the validation certificate and the Accountability Log for the NG arm. For all subsequent treatments, one aliquot will be removed and thawed prior to delivery via the NG or enema route. Sites will be provided with a secondary IMP label for affixing onto individual plastic bags that the FMT aliquots will be placed in to ensure correct administration of treatment route to the correct trial patients. Details to include patient TNO, initials, date of birth, route, treatment number, FMT expiry date and time on day of thawing must be clearly stated on the label prior to attachment and administration.

7.3. Dosing Schedule

Once patients have been randomised into the trial and the treatment arm allocated, the appropriate pre-thawed aliquots of FMT will be available for either administration via the colonoscope or after placement of an NG tube as appropriate.

The FMT will be dispensed as per FMT delivery route allocated:

- NASO-GASTRIC route: 30g of stool in 50ml for naso-gastric administration (1 aliquot) each day for 4 days at the start of the trial (starting on the day of randomisation, week 0) and then again for 4 days in week 4 (see section 7.3.2).

- COLONIC route: 150g of stool in 250ml for colonoscopic administration (5 aliquots) on day of randomisation (week 0) followed by 30g of stool made to 100ml in normal saline for administration via enema each week for 7 weeks (i.e. weeks 1 to 7). At each site, the 50ml aliquots will be made up to 100ml for enema by the addition of 50ml normal saline after thawing of the FMT at room temperature (see section 7.3.3)..

Where possible, Sites should try to contact the patient on the morning of each subsequent treatment visit, prior to thawing the FMT aliquots, to confirm patient attendance and therefore preventing wastage of the aliquot.

7.3.1. Trial-related Drugs

Three routine clinical drugs will be used as adjuncts in this trial alongside routine bowel-preparation methods as outlined in Table 1 below.

Table 1: Dosing regimen

Medication	Dose/Regimen
Bowel-preparation	2 litres of reconstituted Moviprep® solution within the 24 hours prior to procedure – this may be split into two 1 litre doses as needed.
Domperidone (NG arm only)	10mg dose
Lansoprazole (NG arm only)	30mg dose
Loperamide	2mg dose

7.3.2. Naso-gastric FMT

The day before attending for the initial FMT treatment all patients will receive standard bowel-preparation as described in Table 1 above. They will then be colonoscoped.

Patients will be pre-treated with a proton pump inhibitor (lansoprazole) and a prokinetic agent (domperidone) at least 30 minutes before each FMT infusion to reduce gastric secretion and prevent the risk of regurgitation respectively. NG tubes will be passed and checked for correct position as per the local Site protocol for NG tube insertion. Following colonoscopy, 50ml thawed FMT treatment will be infused as per the **STOP-Colitis** protocol. In this NG route, 4x50ml FMT aliquots, i.e. 1x50ml aliquot for each day will be delivered over 4 consecutive days, including the first dose delivered on the day of randomisation. Patients in the NG arm will then have a further 4 days treatment beginning at the week 4 visit following a fast from midnight. Patients will receive single dose loperamide after each FMT delivery.

After each FMT delivery, the NG tube will usually be removed unless patients express that they wish to keep the tube in situ for 4 days, in which case the tube will then be removed at day 4 (or on the last day of their treatment in case of treatment delays) to be replaced for the second course of treatment. Sites must follow their local hospital policy for nasogastric tube care. In the event of NG tubes falling out prior to the completion of the course of treatment, they will be replaced and position checked as per local NG tube insertion policy.

In this NG arm of the pilot study, patients who complete the treatment according to the protocol will receive in total, 240g FMT.

After each FMT delivery, patients may be monitored for up to 6 hours in the Clinical Research Facility. Medically trained doctors will examine patients and in the unlikely event of significant adverse event(s) in relation to colonoscopy or the development of systemic symptoms such as vomiting, pyrexia or hypotension, clinical investigators will consider admitting patients overnight.

7.3.3. FMT by the colonic route

The day before attending for the initial FMT treatment, all patients will receive standard bowel-preparation as described in Table 1 above. On the day of the planned treatment, patients will undergo standard colonoscopy and FMT using a thawed 250ml FMT aliquot in normal saline and 10% glycerol containing 150g of donor stool. Of the 250ml suspension, 125ml will be sprayed into the caecum via a spray catheter at colonoscopy with the remaining 125ml sprayed directly onto the rest of the colon (after samples obtained). Weekly faecal enemas will be administered containing 30g donor stool made up to 100ml with normal saline in 10% glycerol up to week 7. Patients will get a single dose of loperamide after each FMT delivery.

In this COLON arm, patients who complete the treatment according to the protocol will receive in total 360g FMT.

After each FMT delivery, patients may be monitored for up to 6 hours in the Clinical Research Facility. Medically trained personnel will examine patients and in the unlikely event of significant adverse event(s) in relation to colonoscopy or the development of systemic symptoms such as vomiting, pyrexia or hypotension, clinical investigators will consider admitting patients overnight.

7.4. Concomitant drugs

7.4.1. Prohibited drugs

Patients will not be able to take oral or systemic steroids or change their maintenance treatment for UC for the first 8 weeks of the pilot study. However, should a patient develop symptoms of a flare of disease needing additional or incremental treatment (e.g. steroids or antibiotics) during the trial they will be clinically reviewed by a local STOP-Colitis investigator. At this time, assessments will be made and patients who have a clinically significant relapse will be allowed treatment escalation. Participants will be requested to continue to provide scheduled samples as per protocol. This will be considered a trial end point, “treatment failure”; these patients will not continue FMT treatment. Details of treatment escalation will be recorded on the Maintenance Therapy Form.

7.4.2. Allowed drugs

Patients will be able to continue with maintenance medication for UC (e.g. oral 5ASA compounds, thiopurines or methotrexate) if on stable doses for 3 months prior to study entry.

7.5. Treatment modifications

All patients will be encouraged to continue with the study schedule as per protocol irrespective of the total number of treatments they receive. Patients will also be encouraged to continue in the study if they do not tolerate one of their doses due to the development of side-effects which are regarded as clinically mild and self-limiting.

In the event of a treatment delay, the maximum delay allowed between scheduled doses is 2 days for both treatment delivery routes. Where a treatment delay occurs beyond 2 days, the dose will be abandoned and subsequent treatments will continue as per original treatment schedule. All treatment delays will be recorded on the **STOP-Colitis** data collection forms.

If a patient experiences severe side effects (see Section 7.6), the patient will permanently discontinue FMT treatment and will not receive any further FMT treatment.

7.6. Permanent discontinuation of FMT

FMT treatment will be stopped in the event of any of the following being reported during a treatment course:

- New pyrexia ≥38°C
- Severe exacerbation of IBD during or after any FMT treatment course with an increase in partial Mayo score of more than 2 points on the weekly assessment
- Hypotension (drop in systolic blood pressure below 90mmHg)
- Tachycardia (pulse rate ≥100)
- Any other reason or situation (e.g. severe side-effects, SAE) as deemed by the PI to warrant discontinuation of FMT treatment

If a patient experiences any of the above, the patient will permanently discontinue FMT treatment and will not receive any further FMT.

If a patient permanently discontinues FMT, this must be communicated immediately in writing (by e-mail) to the **STOP-Colitis** Trial Office. The site should then complete the Patient Change of Status Form.

7.6.1. Disposal of FMT following patient discontinuation or withdrawal

The **STOP-Colitis** Trial Office will notify the UoBMTC should it be necessary for the UoBMTC to analyse the FMT reference sample that was retained at UoBMTC following QP batch release of the treatment set.

Any FMT aliquots remaining for the patient's treatment course at Site will need to be disposed of as per local clinical waste disposal protocol following instruction from the **STOP-Colitis** Trial Office. The details of disposal of each aliquot must be documented on the FMT Receipt and Accountability Log.

8. TRIAL PROCEDURES AND ASSESSMENTS

8.1. Table 2: Naso-Gastric FMT Schedule of assessments

	Registration & Screening	Randomisation & Treatment start	Wk 1	Wk 2	Wk 3	Wk 4	Wk 5	Wk 6	Wk 7	Wk 8	Wk 12	ETV
Consent	X	X										
Physiological measurements	X	X								X	X	
Inclusion/ Exclusion Criteria	X	X										
IBD Diaries dispensed & collected	X	X	X	X	X	X	X	X	X	X	X	
IBDQ & SF-36		X (prior to randomisation)								X	X	
Qualitative research interview arranged	X										X	
Partial Mayo	X	X	X	X	X	X	X	X	X	X	X	X
Stool for CPT /microbiome and calprotectin arranged	X		X		X		X		X	X		
Stool for CPT /microbiome and calprotectin collected		X		X		X		X		X	X	
Stool for C. diff / enteric pathogens arranged (collected prior to randomisation)	X											
FBC/BIO/CRP	X					X		X		X		
Spot urine test (females)	X											
Dispense Bowel Prep	X											
Urine sample		X								X	X	
Administer Domperidone and Lansoprazole		X				X						
Colonoscopy		X										
Colonic Biopsies		X								X		
Full Mayo Score		X								X		
X-ray (only if necessary)		X				X						
FMT NG		**4 DAYS**				**4 DAYS**						
Administer Loperamide		X				X						
Adverse Events		X	X	X	X	X	X	X	X	X	X	
Medication		X	X	X	X	X	X	X	X	X	X	
Flexible Sigmoidoscopy										X		

Key:

- IBD=Inflammatory Bowel Disease
- IBDQ=Inflammatory Bowel Disease Questionnaire
- SF-36=Short-Form 36
- CPT= calprotectin
- FBC=Full Blood Count- Haemoglobin, WBC, Platelets
- BIO=Biochemical assessment- Urea, ALT, AST, Albumin, Creatinine
- CRP=C-reactive protein
- ETV=Early Termination Visit

8.1.1. NASO-GASTRIC ROUTE

Registration & Screening (-20 to Day 0): Following written informed consent to screening for **STOP-Colitis**, patients will be registered to the trial. They will undergo basic physiological measurements (pulse, blood pressure, temperature, height and weight). Patient inclusion/exclusion criteria will be reviewed and blood tests will be undertaken. They will be questioned about their bowel habits as a rough estimate to calculate the partial Mayo with regard to symptoms on the three days prior to screening. They will receive weekly IBD diaries to record bowel symptoms for the duration of the trial. These diaries will be collected weekly and will remain at the hospital to calculate the partial Mayo score (Appendix 1).

Standard bowel preparation kits (Moviprep®) will also be given to the patient at this screening visit in addition to stool sample collection kits. Patients will be asked to return the *Clostridium difficile* stool sample as soon as possible to the hospital, so that the result is available to confirm eligibility for randomisation. The IBD team will contact the patient to notify them of their stool result and, if tested negative for *Clostridium difficile,* will instruct the patient to take the bowel preparation. Patients should collect a stool sample on the day of bowel preparation, before taking the bowel preparation. The stool sample should be stored in the fridge and brought to the hospital the next day.

The patient will complete the Patient Contact Details Form which will be sent by the IBD team to the **STOP-Colitis** Trial Office for forwarding to the qualitative researcher working with the research team. The qualitative researcher will contact the patients to arrange an initial interview prior to their visit for randomisation and treatment start (see section 8.3)

Randomisation & Treatment start (Day 0): Prior to randomisation, written informed consent will be obtained. Patients will undergo basic physiological measurements (pulse, blood pressure, temperature, height and weight). Blood test results will be checked and IBD diary cards reviewed to calculate the partial Mayo score. The stool sample will be collected. A urine sample will be taken for metabolomics studies. Baseline QoL assessments to include the Inflammatory Bowel Disease Questionnaire (IBDQ) and generic Short-Form 36 (SF-36) will be completed by patients before randomisation. Following confirmation of eligibility and calculation of the partial Mayo score based on diary entries for the 3 days preceding this visit, patients will be randomised to receive FMT via either the NG or COLON route.

For logistical purposes, given the relatively short time between the patient arriving at the Clinical Research Facility and the potential administration of the FMT, it may be necessary, at the start of the day, to thaw 5 x 50ml FMT aliquots for colonoscopic delivery in all cases. If the patient is then randomised to NG, it must be accepted that 4 x 50ml FMT aliquots will be wasted in this case**.**

Once randomised, all patients will have a colonoscopy to assess disease and collect mucosal biopsies. Two biopsies will be taken from the ascending, descending colon and rectum on intubation for later microbiome analysis. At the same time, one biopsy will be taken from these sites for routine histological assessment. Patients randomised to NG will have full colonoscopy for the full Mayo score to be calculated (see below) with biopsy for later microbial studies. They will have a dose of domperidone and lansoprazole following completion of

randomisation. An NG tube will be passed and position checked as per local hospital protocol for NG tube placement and correct positioning. Once it has been confirmed that the tip of the NGT is in the stomach, FMT will be performed as described above. All patients will receive a dose of loperamide after each FMT dose.

A full Mayo score (Appendix 2) will be calculated. Patients will be assessed for disease activity at entry using a combination of the standard Mayo clinical scoring system and surrogate serum (CRP) and stool (calprotectin) markers of colonic inflammation. Patients will be followed up weekly, approximately every 7 days for 8 weeks.

Days 1 to 3 NG FMT Treatment: Patients will return to the Clinical Research Facility daily for 3 days and will continue to receive FMT treatment via the NG tube. Patients will be pre-treated with a dose of domperidone and lansoprazole prior to the FMT infusion and will receive a dose of loperamide after each FMT dose. Information regarding medication use and details of any adverse events occurring will be documented.

Weeks 1-7: Partial Mayo score will be calculated from IBD diaries collected each week. Stool kits for microbiome and calprotectin will be dispensed at weeks 1, 3, 5 and 7 and collected at weeks 2, 4 and 6. At weeks 4 and 6, further blood samples will be taken for CRP assessment. At each weekly visit, information on medication use will be taken and details of any adverse events occurring will be documented.

Week 4 (days 1 to 4) NG FMT Treatment: At the week 4 visit, following a midnight fast, patients will attend the Clinical Research Facility. They will have a dose of domperidone and lansoprazole after which an NG tube will be passed (if tube was removed at week 1) and 30g, 1 x 50ml FMT administered. All patients will then receive a dose of loperamide. This will be repeated for a further 3 days as per the first treatment. Patients have the option of retaining the tube or having it removed after each treatment and replaced daily. Each time the tube is replaced, it will be passed and position checked as per local hospital protocol for NG tube placement and correct positioning.

Week 8: Physiological measurements will be performed. Patient IBD diaries will be collected by the hospital IBD team to calculate the partial Mayo score and new IBD Diaries will be issued. Patients will complete the QoL questionnaires, as completed at randomisation. Stool for calprotectin, stool and urine for metagenomics and metabolomics will be collected. New stool kits will be dispensed for stool collection at week 12. Blood and urine samples will also be taken. Information regarding medication use and details of any adverse events occurring will be documented.

A full Mayo score will be calculated. A flexible sigmoidoscopy will be performed and samples taken for mucosal histological assessment and later mucosal microbiome assessment. Two biopsies will be taken from the descending colon and rectum for microbiome assessment and one from each of these sites for routine histological assessment of inflammation.

Week 12: At this last follow-up visit, physiological measurements will be undertaken, IBD diaries collected and a partial Mayo score calculated. Patients will complete the QoL questionnaires as completed at randomisation and week 8. Stool and urine samples will be collected. Information regarding medication use and details of any late occurring adverse events will be documented.

Patients will take part in a follow-up semi-structured qualitative research interview as soon as possible after this week 12 assessment in a non-clinical setting.

NG Early Termination Visit (ETV)

If patients discontinue trial treatment permanently, they will be invited to continue to attend for scheduled follow up visits. If they decide to withdraw from the trial altogether they will be invited to attend for an early termination visit. At this time only a partial Mayo score will be calculated.

Patients will then be contacted by the qualitative researcher for an interview soon after should they still be willing to be contacted.

8.2. Table 3: COLONIC FMT Schedule of assessments

	Registration & Screening	Randomisation & Treatment start	Wk 1	Wk 2	Wk3	Wk 4	Wk 5	Wk 6	Wk 7	Wk 8	Wk 12	ETV
Consent	X	X										
Physiological measurements	X	X								X	X	
Inclusion/ Exclusion Criteria	X	X										
IBD Diaries dispensed and collected	X	X	X	X	X	X	X	X	X	X	X	
IBDQ & SF-36		X (prior to randomisation)								X	X	
Qualitative research – interview arranged	X										X	
Partial Mayo score	X	X	X	X	X	X	X	X	X	X	X	X
Stool for CPT /microbiome and calprotectin arranged	X		X		X		X		X	X		
Stool for CPT /microbiome and calprotectin collected		X		X		X		X		X	X	
Stool for C. diff / enteric pathogens arranged (collected prior to randomisation)	X											
FBC/BIO/CRP	X					X		X		X		
Spot urine test (females)	X											
Dispense Bowel Prep	X											
Urine sample		X								X	X	
Colonoscopy		X										
Colonic Biopsies		X								X		
Full Mayo		X								X		
FMT COLON		**X**	**X**	**X**	**X**	**X**	**X**	**X**	**X**			
Administer Loperamide		X	X	X	X	X	X	X	X			
Adverse Events		X	X	X	X	X	X	X	X	X	X	
Medication		X	X	X	X	X	X	X	X	X	X	
Flexible Sigmoidoscopy										X		

Key:

- IBD=Inflammatory Bowel Disease
- IBDQ=Inflammatory Bowel Disease Questionnaire
- SF-36=Short-Form 36
- CPT= calprotectin
- FBC=Full Blood Count- Haemoglobin, WBC, Platelets
- BIO=Biochemical assessment- Urea, ALT, AST, Albumin, Creatinine
- CRP=C-reactive protein
- ETV=Early Termination Visit

8.2.1. COLONIC FMT

Registration & Screening (-20 to Day 0): Following written informed consent to screening for **STOP-Colitis**, patients will be registered to the trial. They will undergo basic physiological measurements (pulse, blood pressure, temperature, height and weight). Patient inclusion/exclusion criteria will be reviewed and blood tests will be undertaken. They will be questioned about their bowel habits as a rough estimate to calculate partial Mayo with regard to symptoms on the three days prior to screening. They will receive weekly IBD diaries to record bowel symptoms for the duration of the trial. These diaries will be collected weekly and will remain at the hospital to calculate the partial Mayo score (Appendix 1).

Standard bowel preparation kits (Moviprep®) will also be given to the patient at this screening visit in addition to stool sample collection kits. Patients will be asked to return the *Clostridium difficile* stool sample as soon as possible to the hospital, so that the result is available to confirm eligibility for randomisation. The IBD team will contact the patient to notify them of their stool result and, if tested negative for *Clostridium difficile,* will instruct the patient to take the bowel preparation. Patients should collect a stool sample on the day of bowel preparation, before taking the bowel preparation. The stool sample should be stored in the fridge and brought to the hospital the next day.

Patients will complete the Patient Contact Details Form which will be sent by the IBD team to the **STOP-Colitis** Trial Office for forwarding to the qualitative researcher working with the research team. The qualitative researcher will contact the patients to arrange an initial interview prior to their visit for randomisation and treatment start (see section 8.3)

Randomisation & Treatment start (Day 0): Prior to randomisation, written informed consent will be obtained. Patients will undergo basic physiological measurements (pulse, blood pressure, temperature, height and weight). Blood test results will be checked and IBD diary cards reviewed to calculate the partial Mayo scores. The stool sample will be collected. A urine sample will be taken for metabolomics studies. Baseline QoL assessment to include, the Inflammatory Bowel Disease Questionnaire (IBDQ) and generic Short-Form 36 (SF-36) will be completed by patients before randomisation. Following confirmation of eligibility and calculation of the partial Mayo score as based on diary entries for the 3 days preceding this visit, patients will be randomised to receive FMT via either the NG or COLON route.

For logistical purposes, given the relatively short time between the patient arriving at the Clinical Research Facility and the potential administration of the FMT, it may be necessary, at the start of the day to thaw 5 x 50ml FMT aliquots for colonoscopic delivery in all cases.

Once randomised, all patients will undergo colonoscopy to assess disease and calculate full Mayo score (see below), collect mucosal biopsies on intubation and receive the first dose of FMT via the endoscope. All patients will receive a dose of loperamide after each FMT dose. Two biopsies will be taken from the ascending, descending colon and rectum for later microbiome analysis. At the same time, one biopsy will be taken from these sites for routine histological assessment.

A full Mayo score (Appendix 2) will be calculated. Patients will be assessed for disease activity at entry using a combination of the standard Mayo clinical scoring system and surrogate serum (CRP) and stool (calprotectin) markers of colonic inflammation. Patients will be followed up weekly, approximately every 7 days for 8 weeks.

Weeks 1-7 COLON FMT Treatment: Patients will return to the Clinical Research Facility weekly and will receive a dose of FMT treatment by enema for 7 weeks followed by loperamide. Further IBD diaries will be dispensed and collected at each week to calculate partial Mayo scores. Stool kits for microbiome and calprotectin will be dispensed at weeks 1, 3, 5 and 7 and collected at weeks 2, 4 and 6. At weeks 4 and 6, further blood samples will be taken for CRP assessment. At each weekly visit, information on medication use will be taken and details of any adverse events occurring will be documented.

Week 8: Physiological measurements will be performed. Patient IBD diaries will be collected by the hospital IBD team to calculate the partial Mayo scores and new IBD diaries will be issued. Patients will complete the QoL questionnaires, as completed at randomisation. Stool for calprotectin, stool and urine for metagenomics and metabolomics will be collected. New stool kits will be dispensed for stool collection at week 12. Blood samples will also be taken. Information regarding medication use and details of any adverse events occurring will be documented.

A full Mayo score will be calculated. A flexible sigmoidoscopy will be performed and samples taken for mucosal histological assessment and later mucosal microbiome assessment. Two biopsies will be taken from the descending colon and rectum for microbiome assessment and one from each of these sites for routine histological assessment of inflammation.

Week 12: At this last follow-up visit, physiological measurements will be undertaken, IBD diaries collected and a partial Mayo score calculated. Patients will complete the QoL questionnaires as completed at randomisation and week 8. Stool and urine samples will be collected. Information regarding medication use and details of any late occurring adverse events will be documented.

Patients will take part in a follow-up semi-structured qualitative research interview as soon as possible after this week 12 assessment in a non-clinical setting.

COLON Early Termination Visit (ETV)

If patients discontinue the trial treatment permanently, they will be invited to continue to attend for scheduled follow up visits. If they decide to withdraw from the trial altogether they will be invited to attend for an early termination visit. At this time only a partial Mayo score will be calculated.

Patients will then be contacted by the qualitative researcher for an interview soon after should they still be willing to be contacted.

8.3. Qualitative Research Interview:

Patient and clinician experience and acceptability of FMT and trial processes

The qualitative research interviewer will conduct research interviews with patients participating in both arms of the trial. Following consent, and registration, each patient will take part in a semi-structured interview at two time points; the first (T1) following the screening visit, and prior to randomisation and the second (T2) will take place soon after completion of the 12 week follow up visit. These one-to-one interviews with the qualitative research fellow will be conducted either in person, over the telephone or by Skype as preferred by the participants. T1 interviews will include a Background Questionnaire to be filled in by the patients. Information on age, ethnicity, education, employment and some general questions around the patient's UC will be collected on the Background Questionnaire. During the T1 interview, patients' history of UC and treatment to date, and also their understanding and expectations for FMT and the pilot study will be explored. During the T2 interviews, the patients' actual experience within the pilot study, including that of FMT (by NG and COLON) and of related trial processes and procedures will be explored. Interviews will also gather data on patient perspectives

regarding important impacts and outcomes of FMT, thus providing the opportunity to compare these with the outcome measures selected for inclusion in the pilot and (later) full RCT.

Further interviews with patients who choose to withdraw from the trial early and with a small sample of patients who decline consent to participate following initial screening for eligibility (should this happen), will provide in-depth understanding of these decisions. Interviews with patients who choose not to take part in the study will be conducted by the qualitative researcher, either in person or over the telephone, as preferred by the participants. These interviews will include a Background Questionnaire, as described above, and will give insight into their reasons for choosing not take part in the study.

Interviews will also be conducted with staff at the pilot sites to review the experience of trial conduct e.g. the acceptability and logistics of complex trial procedures within clinical environments. Data from this qualitative research will contribute to an assessment of patient and clinician experience and acceptability of FMT within the pilot context, and along with recruitment and drop-out rates, this will inform a decision about whether the NG or COLON route of delivery is taken forward to the full RCT. The qualitative data will also contribute to the refinement and optimisation of trial processes prior to the full RCT, and allow construction of survey tools which can be used to assess patient and clinical acceptability within the full trial.

Analysis of qualitative data: Interviews will be recorded with the consent of participants and transcribed clean verbatim for analysis. Analysis will be conducted with reference to recordings, transcripts and field notes taken at the time of data collection. A thematic analysis of content will be informed by the Framework analytical approach. Following initial familiarisation with the interview data, development of thematic frameworks and data coding will proceed in an iterative manner. Data collection and analysis will run concurrently so that emergent analytical themes can inform further data collection, and particularly comparative analytical questioning between patients allocated to NG or COLON.

8.4. Partial and full Mayo scoring

The Mayo Scoring system is the most commonly used system for assessment of UC activity. Partial Mayo scores will be calculated using the **STOP-Colitis** IBD diaries provided with regard to symptoms collected 3 days preceding each weekly hospital visit. Please refer to Appendix 1 for the clinical scoring table for the partial Mayo and Appendix 2 for the full Mayo.

8.5. Quality of life assessments

Two validated patient completed questionnaires will be used as an assessment of quality of life. These include the IBDQ as licensed for use by McMaster University and the SF-36 v2 licensed for use by OPTUM.

All questionnaires will be provided to sites as part of the ISF. It will be the responsibility of the IBD research team to provide the questionnaires to patients recruited to **STOP-Colitis**, for completion as per protocol scheduled time-points. Questionnaires must be completed by patients in hospital on the day of the clinic visit. The IBD research team will return all original completed questionnaires via post to the **STOP-Colitis** Trial Office in a timely manner.

8.6. Mechanistic Sub studies

Table 4: Summary of sample collection and dispatch

Sample to be dispatched	Quantities	Timelines	Recipient
Donor urine	10-20ml	Day 1 and 10	University of Glasgow
Donor stool (fresh and frozen)	Approx 10-20g	Day 1 and 10	University of Glasgow
Donor stool (fresh and frozen)	Approx 10-20g	Day 1 and 10	University of Birmingham
Patient stool for calprotectin	Approx 10-20g	Randomisation visit, weeks 2, 4, 6, 8 and 12	University of Glasgow
Patient urine for SCFA	10-20ml	Randomisation visit, weeks 8 and 12	University of Glasgow
Patient stool for SCFA	Approx 10-20g	Randomisation visit, weeks 2, 4, 6, 8 and 12	University of Glasgow
Patient mucosal biopsies	6 biopsies	Randomisation visit, week 8	University of Birmingham
Patient stool for 16SRNA	Approx 10-20g	Randomisation visit, weeks 2, 4, 6 8 and 12	University of Birmingham

8.7. CRP

This will be measured in NHS hospital laboratories using standard methodology.

8.8. Faecal calprotectin

Measurements of faecal calprotectin will be performed using a standard protocol [15] with commercially available ELISA kits and according to the specifications of the manufacturer. Briefly, 100 mg of thawed stool sample will be extracted with a proprietary buffer, then diluted and loaded on a 96 well plate coated with calprotectin antibodies. The concentration of faecal calprotectin will be calculated in mg/kg of wet and dry faecal matter. Faecal extracts will be assayed in duplicates.

As measurements vary according to the laboratory and kits used, all measurements will be performed in one central laboratory in Glasgow. Faecal samples will therefore be dispatched directly to Glasgow.

8.9. Metagenomics

The study will combine 16S rRNA sequencing and profiling of SCFAs of all samples with shotgun metagenomics and full metabolomics on selected samples. In the pilot, 16sRNA sequencing will be undertaken on the following samples:

- Stool from the donor (fresh and frozen from day 1 and day 10 donations)
- Stool from the recipient at randomisation visit, weeks 2, 4, 6, 8 and 12
- Mucosal biopsies taken from right colon, left colon and rectum at randomisation and week 8
- Technical replicates (~15 samples per patient)

DNA for metagenomics from stool and biopsies will be extracted at the University of Birmingham. A 2-stage broad and deep experimental design will be used for the study of the microbiome in UC on patient samples collected before, during and after faecal transplantation employing cutting-edge metagenomics analysis techniques. We will apply conventional phylogenetic profiling techniques using deep sequencing of 16S ribosomal RNA marker gene to all samples, both at multiple time points for each patient and the initial donor stool sample.

8.10. Major bacteria metabolites and metabolomics

Major bacterial metabolites previously implicated in the aetiology and mucosal inflammation of IBD will be measured with GC-FID/MS and other assays as described previously. Metabolites will include faecal short and medium chain fatty acids (C2-C8), branch chain fatty acids (iC4-iC6), lactate, ammonia, sulphide, phenols and

cresols and pH[k1]. These metabolites will be measured at all time points of the study where stool collection is taking place. Data will be presented in absolute concentrations per dry matter and relative ratios (%).

8.11. Urinary and faecal metabolomics

Samples for analysis will include:
(SCFA only):

If not proceeding to RCT, SCFA and 16sRNA sequencing on:

- urine and stool from 30 patients at 0, 8 and 12 weeks
- urine and stool from donors on day 1 and day 10

If proceeding to the RCT. SCFA, 16sRNA sequencing and metabolomics on all the above.

Faecal and urinary samples will be analysed by the Glasgow Polyomics Facility, University of Glasgow http://www.polyomics.gla.ac.uk/ which is equipped with LC-MS (Orbitrap Exactive, and Orbitrap Velos), GC-MS and NMR capability for collecting metabolomics data. Analysis will be carried out in-house using pipelines that provide robust metabolite identification and quantification algorithms. We will collect metabolomics data using well-established methodologies [30-33].Core metabolite identifications will be validated against a panel of authentic standards by mass and retention time, and additional standards may be used to confirm novel metabolites of significant interest. Where necessary, confirmation of metabolite identity will be made using fragmentation and comparison to an authentic standard. Very volatile compounds (e.g. free sulphides and ammonia) which will not stand storage and transport will be analysed on site locally by the research fellow running the study.

Measurement of SCFA

Thawed samples will be fixed 1:1 v/v with 1M NaOH and will subsequently be freeze-dried by lyophilization. 100 mg of freeze-dried material will be extracted three times with diethyl ether in acidified aqueous samples. The ether extracts will be analysed on a Gas Chromatographer with Flame Ionization Detector. Absolute and relative quantification will be performed against authentic internal and external standards of fatty acids from C2 to C8 plus the branched chain fatty acids iso-butyrate, iso-valerate and iso-hexanoic. Each sample will be extracted in double and the average concentration will be calculated unless the %CV is more than 10% in which case a third extraction will be performed.

8.12. Donor Assessment of dietary intake

The habitual dietary pattern of the donors will be assessed using the validated food frequency questionnaire (FFQ) as used in the EPIC study in the UK [29]. Instructions on how to complete the FFQ will be provided to the donors by research nurses at the Clinical Research Facility. Data from the FFQ questionnaires will be transferred to Glasgow for analysis. Energy, macronutrient and fibre intake will be estimated and expressed in nutrient ranks and quartiles and will also be compared against the DoH recommendations and the UK NDNS survey results.

8.13. Bioinformatics and Statistical Analysis

Amplicon sequence data will be analysed using the QIIME pipeline including steps for filtering, chimera removal, de novo OTU prediction and phylotyping with taxonomic assignments against the SILVA database. This will provide us with a taxonomic profile for each sample, which will be coupled with results of the major metabolite analysis. Together these samples will be compared to clinical data, and correlations found with faecal calprotectin, in order to find the samples most likely to be biologically informative as a marker of disease remission. The entire DNA set will be co-assembled using Ray on our high-memory (3Tb) machines on the Medical Research Council (MRC) funded Cloud Infrastructure for Microbial Bioinformatics (CLIMB) and genome sequences reconstructed using the applicant's CONCOCT software [39]. Genomes will be annotated, and functional assignments made against the EggNog and KEGG databases. Multivariate analysis will be employed to detect clusters associated with recovery and relapse, and genome-level information will be used to

reconstruct strains in the input pool which persist or are lost in recovery or relapse to inform a mechanistic understanding of the role of bacterial strains in faecal transplantation. This will enable us to correlate the metabolic signatures directly with the functional profiles and the annotated genome abundances and, hence, determine the metabolic roles of the key community members.

8.14. Microbiota culture from donor samples

Fresh faecal samples from donors taking part in the **STOP-Colitis** trial will be collected and transferred to sterile specimen containers immediately following defecation. These will then be stored anaerobically in airtight bags containing an anaerobic pouch (AnaeroGen Compact, Oxoid, UK) and an ice-pack until transfer to an anaerobic chamber within 1–2 hours of collection.

0.25 mg of faecal material will be homogenised in 1 ml PBS using bead tubes (CK01, 0.1 mm ceramic beads, Precellys). Samples will then be centrifuged for 1 minute at 10,000g, followed by serial dilution in peptone water. These will then be cultured using a variety of general purpose and selective media (Yeast Casitone Fatty Acid (YCFA) Agar, Brain Heart Infusion (BHI) Agar, Reinforced Clostridial Agar, Wilkins Chalgren Agar, Fastidious Anaerobe Agar, Rogosa Agar, Perfringens OPSP, CCFA, Beeren/Bifidobacterium Agar, Blood Azide Agar, McConkey Agar and Nutrient Agar) for the culture of aerobes and fastidious anaerobes. The plates will then be incubated aerobically and anaerobically at 37°C. Bacterial colonies will be streaked to purity and single colonies cultured in relevant specific media and create pure frozen stocks.
From each frozen stock, cultures will be grown and used for colony identification at the species level through polymerase chain reaction (PCR) product sequencing of the 16S rRNA gene using the universal primers 8F (5'-AGAGTTTGATCCTGGCTCAG-3') and 1492R (5'-ACGGCTACCTTGTTACGACTT-3'). Bacterial genomic DNA from each culture will be extracted using the Bacterial Genomic Miniprep Kit (Sigma Aldrich). PCR will then be carried out in 25-µL reactions, containing 10 mM Tris·HCl (pH 8.3), 50 mM KCl, 1.5 mM MgCl2, 200 µM of each dNTP, 0.5 U Taq DNA polymerase (Takara, Shiga, Japan), 0.4 µM of each respective primer, and 10 ng DNA template. Specific PCR conditions will be applied and amplifications performed on a Thermal Cycler. Amplicons will be purified and analysed using a Tapestation (Agilent Technologies) to detect primer-dimers and determine average molecular weight of each product. The PCR product will then be cleaned up using a PCR clean up kit (Wizard, Promega). Nucleotide sequences will then be determined by the dideoxynucleotide method using cycle sequencing (Sanger sequencing) (Source BioScience). Sequences will be subjected to BLAST searches within the GenBank database (http://www.ncbi.nlm.nih.gov/) to determine 16S rRNA gene sequence similarities to cultured and not yet cultured organisms.

Up to 30 isolates will be selected in the first instance based on the 16s rRNA Sanger sequencing results and whole genome sequencing will be performed on these to obtain strain level identification. Additionally purified cultures will be verified using matrix assisted laser desorption ionization-time of flight mass spectronomy (MALDI-TOF), using previously published protocols [Nagy *et al.*, 2009].

8.15. Patient withdrawal and Change of Status within the Trial

Participants should be made aware at the beginning of study entry that they can freely withdraw (discontinue participation) from the trial at any time.

A participant who wishes to cease to participate in a particular aspect of the trial, will be considered as having changed their status within the trial.

The changes in status within the **STOP-Colitis** trial are categorised in the following ways:

- **No trial intervention:** The participant would no longer like to receive the trial intervention, i.e. permanently discontinues FMT treatment but is willing to be followed up in accordance with the schedule of assessments and if applicable using any central UK NHS bodies for long-term outcomes (i.e. the participant has agreed that data can be collected and used in the trial analysis).

- For patients who discontinue treatment, clinic visits and follow-up data will continue to be collected until the last visit at week 12. A post 12-week qualitative research interview will be arranged with the patient.

- **No trial intervention AND no further data use:** The participant would like to discontinue from FMT treatment, is not willing to be followed up in any way for the purposes of the trial and does not wish for any further data to be collected (i.e. only data collected prior to the withdrawal can be used in the trial analysis). This will be regarded as an Early Termination from the trial.

- **The participant would like to withdraw from qualitative research interviews only:** The participant is willing to continue FMT treatment and follow-up visits as per the protocol but does not wish to participate in the qualitative interview at Week 12.

The details of change of status within trial (date, reason and category of status change) should be clearly documented in the source data and on a **STOP-Colitis** Patient Change of Status Form.

In any case of withdrawal, patients will be offered standard care as per local hospital protocol for UC management.

9. PHARMACOVIGILANCE

9.1. Adverse Events Reporting Requirements

The collection and reporting of all Adverse Events (AEs) within **STOP-Colitis** will be in accordance with the Medicines for Human Use Clinical Trial Regulations 2004 and its subsequent amendments as applicable. Definitions of different types of AEs are listed in the table of abbreviations and definitions. The PI will assess the seriousness and causality (relatedness) of all AEs experienced by the trial participant.

9.2. Adverse Events

AEs are uncommonly encountered in participants receiving FMT which is NICE-approved for the treatment of recurrent *Clostridium difficile* associated colitis. However, FMT has rarely been used for UC in a clinical trial setting and therefore all AEs experienced whether during or after treatment will be recorded on the **STOP-Colitis** case report forms by research staff at the recruiting Sites. The following expected AEs, as identified through current literature, may occur in trial participants:

- Bloating (primarily due to bowel preparation)
- Transient changes to bowel habits
- Constipation
- Diarrhoea
- Nausea
- Vomiting
- Transient pyrexia
- Epistaxis (due to NG tube)
- Abdominal pain
- AEs clearly associated with the use of standard immune-suppressive medications that patients may be on as maintenance treatment for UC

These AEs are self-limiting but may require symptomatic treatment. It is recognised that these AEs are easily manageable.

9.2.1. AE Reporting period

AEs will be collected during and after each FMT treatment and then at each weekly visit, and at the week 8 and 12 visits. All AEs will be documented and recorded on the case report form relevant to that time point and assessed for casualty and severity by the Investigator.

9.3. Serious Adverse Event Reporting Requirements

Investigators will report AEs that meet the definition of a SAE, other than the SAEs relating to hospitalisation as defined in section 9.3.1

An SAE Form will be completed for these events and will be faxed to the **STOP-Colitis** Trial Office immediately and no later than 24 hours of the site becoming aware of the SAE.

An SAE is defined as an untoward event which:

- Is fatal or immediately life threatening
- Requires or prolongs hospitalisation
- Results in persistent or significant disability or incapacity
- Constitutes a congenital anomaly or a birth defect
- May jeopardise the patient and may require medical or surgical intervention to prevent one of the outcomes listed above

9.3.1. Events that do not require reporting on a Serious Adverse Event Form

For the purposes of **STOP-Colitis**, the following hospitalisations are NOT considered as SAEs:

- Routine treatment or monitoring of the studied indication, not associated with any deterioration in condition or trial procedures
- Treatment, which was elective or pre-planned, for a pre-existing condition that is unrelated to the indication under study, and did not worsen
- Admission to a hospital or other institution for general care, not associated with any deterioration in condition or trial procedures

9.3.2. Monitoring pregnancies for potential Serious Adverse Events

In the event that a participant or their partner becomes pregnant during the SAE reporting period (see section 9.4), a **STOP-Colitis** Pregnancy Notification Form will be completed by the Site (providing the participant's details) and returned immediately to the **STOP-Colitis** Trial Office.

- If it is the participant who is pregnant, outcome data will be provided on a follow-up pregnancy notification form. The participant will be withdrawn from trial treatment, but not from follow-up. Follow-up will be as per protocol and will continue for an additional 6 months after childbirth.
- Where it is the male participant's partner who is pregnant, consent must first be obtained via the pregnancy release of information form. The participant should be given the pregnancy release of information form to give to their partner. If the partner is happy to provide information on the outcome of their pregnancy they should sign the pregnancy release of information form.

Once consent has been obtained, details of the outcome of the pregnancy will be provided on a follow-up Pregnancy Notification Form. If an abortion, miscarriage, congenital abnormality or birth defect, still birth, or neonatal death is observed, then an SAE Form must also be completed.

9.4. SAE Reporting period

9.4.1. Reporting Procedure - Investigator Sites

Details of all SAEs (except those listed above) will be documented and reported from the date of patient Screening consent to **STOP-Colitis** and until 30 days after the administration of the last FMT treatment.

AEs defined as serious and which require reporting as an SAE will be reported on the **STOP-Colitis** SAE Form. When completing the SAE form, the Clinical Investigator will be assess the causality and the severity of the AE.

The following categories will be used to define the relatedness (causality) of the SAE:

Category	Definition	Causality
Definitely	There is clear evidence to suggest a causal relationship, and other possible contributing factors can be ruled out	Related
Probably	There is evidence to suggest a causal relationship, and the influence of other factors is unlikely	
Possibly	There is some evidence to suggest a causal relationship, however, the influence of other factors may have contributed to the event (e.g. the patient's clinical condition, other concomitant events or medication)	
Unlikely	There is little evidence to suggest there is a causal relationship; there is another reasonable explanation for the event (e.g. the patient's clinical condition, other concomitant events or medication)	Unrelated
Not related	There is no evidence of any causal relationship	

On becoming aware that a participant has experienced an SAE, the Clinical Investigator (or delegate) must complete an SAE form providing as much detail as possible and then clearly date and sign the form. The form should accompany an SAE Fax Cover Sheet and arrive at the **STOP-Colitis** Trial Office at BCTU by fax or e-mail immediately and within 24 hours after the Investigator (or delegate) first became aware of the event:

ALL SAES must be recorded on the SAE form and faxed or e-mailed to the STOP-Colitis Trial Office at BCTU on:

0121 415 8871 OR 0121 415 9136

stop-colitis@trials.bham.ac.uk

within 24 hours of becoming aware of the event

On receipt of the SAE form, the **STOP-Colitis** Trial Co-ordinator will allocate each SAE a unique reference number and will also confirm receipt of the SAE and the SAE Reference Number back to the reporting Site. If confirmation of receipt is not received within 1 working day, site research staff must contact the **STOP-Colitis** Trial Office.

For SAE forms completed by someone other than the Investigator, the Investigator will be required to countersign the original SAE form to confirm agreement with the causality and severity assessments. The form should then be returned to the **STOP-Colitis** Trial Office and a copy kept in the ISF.

Investigators should also report SAEs to their own Trust in accordance with local practice.

9.4.2. Provision of follow-up information

Participants will be followed up until resolution or stabilisation of the event. Follow-up information should be provided on a new SAE Form with the follow-up box clearly marked to indicate that this is a follow-up SAE.

9.5. Reporting Procedure – Trials Office

On receipt of an SAE form, the **STOP-Colitis** Trial Office will allocate each SAE a unique reference number. This SAE reference number will be quoted on all correspondence and follow-up reports regarding the SAE and filed with the actual SAE in the Trial Master File. Follow-up SAEs with not be allocated another SAE Reference Number, rather the original SAE Reference number will still apply.

On receipt of an SAE Form, the Chief Investigator (CI) (or a delegate) will assess seriousness and causality independently. An SAE judged by the Investigator or CI to have a reasonable causal relationship with the trial treatment will be regarded as a Serious Adverse Reaction (SAR). The CI, on behalf of the sponsor, will assess all SARs for expectedness. If the event meets the definition of a SAR that is unexpected, i.e. is not defined in the FMT Reference Safety Information (RSI developed through available FMT literature and any future updates; updates as prompted through new available literature on FMT), the event will be classified as an unexpected and related serious adverse reaction. All unexpected and related serious adverse reactions will be dealt with as described in Section 9.6.1.

The Patient Change of Status Form must also be completed for all patients that have been discontinued treatment due to a SAE occurring.

9.6. Reporting to the Competent Authority and Research Ethics Committee

9.6.1. Suspected Unexpected Serious Adverse Reactions

The **STOP-Colitis** Trials Office will report a minimal data set of all individual events categorised as a fatal or life threatening Suspected Unexpected Serious Adverse Reactions (SUSARs) to the Medicines and Healthcare products Regulatory Agency (MHRA) and Research Ethics Committee (REC) within 7 days of receipt of the SAE form. Detailed follow-up information will be provided within an additional 8 days. All other events categorised as SUSARs will be reported within 15 days.

A copy will also be sent to the University of Birmingham Research Governance Team at the time of sending the SUSAR report to the MHRA and REC.

9.6.2. Serious Adverse Reactions

The **STOP-Colitis** Trial Office will report details of all SAEs and SARs (including SUSARs) to the MHRA and REC annually from the date of the Clinical Trial Authorisation, in the form of a Development Safety Update Report (DSUR).

A copy will also be sent to the University of Birmingham Research Governance Team at the time of sending out the DSUR.

9.6.3. Adverse Events

Details of all AEs will be reported to the MHRA on request.

9.6.4. Other safety issues identified during the course of the trial

The MHRA and REC will be notified immediately if a significant safety issue is identified during the course of the pilot trial.

The University of Birmingham Research Governance Team will also be informed at the time that the REC and MHRA is informed.

9.7. Reporting to Principal Investigators

Details of all SUSARs and any other safety issues which arise during the course of the trial will be reported to Principal Investigators who will be responsible for relaying safety information to their IBD research team. A copy of any such correspondence should be filed in the **STOP-Colitis** ISF and reported to the Trust according to local practice.

9.8. Independent Oversight Committee

The Independent Oversight Committee (IOC) will review all SAEs reported in the **STOP-Colitis** Trial regardless of relatedness.

10. DATA HANDLING AND RECORD KEEPING

10.1. Source Data

In order to allow for the accurate reconstruction of the trial and clinical management of the subject, source data will be accessible and maintained at the Sites.

Source data is kept as part of the participants' medical notes generated and maintained at Site. In addition, for **STOP-Colitis** as there is a requirement for endoscopies to be performed, the source data from the endoscopy will be kept in electronic endoscopy reporting programs as used at each of the trial Sites. The IBD Diaries will also be regarded as source data and all patient completed diaries will remain at Site in the ISF. In addition, for this study, patient completed Quality of Life questionnaires will also regarded as source data. This source data will be kept in a locked filing cabinet at BCTU. Copies of the completed questionnaires will also be retained at Sites in the ISF.

10.2. Case Report Form Completion

Data reported on each Case Report Form (CRF) will be consistent with the source data and any discrepancies will be explained. Staff delegated to complete CRFs will be trained to adhere to requirements of data capture as per protocol, regarding for example:

- *Date format and partial dates*
- *Time format and unknown times*
- *Rounding conventions*
- *Trial-specific interpretation of data fields*
- *Entry requirements for concomitant medications(generic or brand names)*
- *Which forms to complete and when*
- *What to do in certain scenarios, for example when a subject withdraws from the trial*
- *Missing/incomplete/unknown data*
- *Completing SAE forms and reporting SAEs*
- *Repeat laboratory tests*

- *Protocol and GCP non-compliances (making amendments)*

In all cases, it remains the responsibility of the site's Principal Investigator to ensure that the CRF has been completed correctly and that the data are accurate.

In **STOP-Colitis**, paper CRFs are used. The completed originals will be submitted to the **STOP-Colitis** Trial Office for entry on the study database, and a copy must be retained in the ISF.

10.3. Data Management

10.3.1. Data collection

All data will be handled in accordance with the General Data Protection Regulation (GDPR) (EU) 2016/679. It will be the responsibility of the investigator to ensure the accuracy of all data entered in the CRFs. The **STOP-Colitis** Signature & Delegation Log will identify all those personnel with responsibilities for data collection.

10.3.2. Qualitative data collection

Interview audio recordings will be held securely on encrypted and password protected computers and networks. Recordings will be transcribed by professional transcription services that have existing data confidentiality agreements with the University of Birmingham, and appropriate data security arrangements. Data will be marked with a unique study ID. All personal identifiers will be removed from hard copy interview transcripts. All qualitative data will only be accessed by the members of the research team.

10.3.3. Data handling and analysis

Paper CRFs must be completed, signed, dated and returned to the **STOP-Colitis** Trial Office by the Investigator or an authorised member of the site research team (as delegated on the **STOP-Colitis** Signature & Delegation Log) within a week of completion. All paper CRFs must also be countersigned by the Principal Investigator or named delegate prior to return.

Entries on the CRF should be made in ballpoint pen, preferably in black ink, and must be legible. Any errors or amendments to data already recorded on a CRF should be crossed out with a single stroke, the correction inserted and each error and/or change initialled and dated. If it is not obvious why a change has been made, an explanation should be written next to the change.

Data reported on each CRF should be consistent with the source data or the discrepancies should be explained. If information is not known, this must be clearly indicated on the CRF. All missing and ambiguous data will be returned for querying with the Site. All sections of the CRFS must be completed in full unless otherwise stated on the CRF.

In all cases, it remains the responsibility of the Investigator to ensure that the CRF has been completed correctly and that the data provided are accurate.

If necessary, CRFs may be amended by the **STOP-Colitis** Trial Office, as appropriate, throughout the duration of the trial. Whilst this will not constitute a protocol amendment, new versions of the CRFs must be implemented by participating sites immediately on receipt.

10.4. Archiving

All records created by following trial procedures and all documents listed in guidance relating to the conduct of the trial must be retained and archived. Archiving will be authorised by the BCTU on behalf of the Sponsor following submission of the end of trial report. It is the responsibility of the Local Principal Investigator to ensure

all essential trial documentation and source documents (e.g. signed Informed Consent Forms, Investigator Site Files, participants' hospital notes, copies of CRFs etc.) at their site are securely retained for at least 25 years.

No documents will be destroyed without prior approval from the **STOP-Colitis** Trials Office.

11. QUALITY CONTROL AND QUALITY ASSURANCE

11.1. Site Set-up and Initiation

Prior to recruitment, all participating Principal Investigators will be asked to sign a Clinical Study Site Agreement and supply a current signed and dated CV and evidence of GCP training to the **STOP-Colitis** Trial Office. All members of the site research team will also be required to sign the **STOP-Colitis** Site Signature and Delegation Log. All sites will undergo a process of initiation and will have completed GCP training prior to activation. Key members of the site research team will be required to attend either a meeting or a teleconference covering aspects of the trial design, protocol procedures, Adverse Event reporting, collection and reporting of data and record keeping. Each Site will be provided with an ISF containing essential documentation, instructions, guidance, SOPs and other documentation required for the conduct of the trial and associated FMT administration documents to be used at each site to record every FMT treatment delivered to patients. The **STOP-Colitis** Trial Office should be informed immediately of any change in the site research team.

11.2. Monitoring

Monitoring will be carried out as required following a risk assessment and as documented in the **STOP-Colitis** Monitoring Plan. **STOP-Colitis** will be centrally monitored, however on-site monitoring may occur if triggered.

The **STOP-Colitis** Trial Office will be in regular contact with the site research teams to check on progress and address any queries that they may have. The **STOP-Colitis** Trial Office will check incoming CRFs for compliance with the protocol, data consistency, missing data and timing. Sites will be asked for missing data or clarification of inconsistencies or discrepancies. Sites will be requested to send the **STOP-Colitis** Trial Office copies of signed Informed Consent Forms and other documentation for in-house review for all participants who have provided explicit consent. This will be detailed in the Monitoring Plan.

On-site monitoring visits may be triggered, for example by poor CRF return, poor data quality, excessive number of participant withdrawals or deviations. If a monitoring visit is required, the **STOP-Colitis** Trial Office will contact the site to arrange a date for the proposed visit and will provide the site with written confirmation. Investigators will allow the **STOP-Colitis** trial staff access to source documents as requested.

11.3. Audit and Inspection

The Principal Investigator will permit trial-related monitoring, quality checks, audits, ethical reviews, and regulatory inspection(s) at their site, providing direct access to source data/documents. The Principal Investigator will comply with these visits and any required follow up. Sites are also requested to notify the **STOP-Colitis** Trial Office of any MHRA inspections.

11.4. Notification of Serious Breaches

The Sponsor is responsible for notifying the REC of any serious breach of the conditions and principles of GCP in connection with that trial or the protocol relating to that trial. Sites must therefore, immediately notify the **STOP-Colitis** Trial Office if any suspected trial-related serious breach of GCP and/or the trial protocol have been identified. Where the **STOP-Colitis** Trial Office is investigating whether or not a serious breach has occurred,

sites must cooperate with the Trial Office in providing sufficient information to report the breach to the REC where required and in undertaking any corrective and/or preventive action.

Sites may be suspended from further recruitment in the event of serious and persistent non-compliance with the protocol and/or GCP, and/or poor recruitment. Any major problems identified during monitoring may be reported to the Trial Management Group and the REC. This includes reporting serious breaches of GCP and/or the trial protocol to the REC. A copy of the serious breach report will be sent to the University of Birmingham Clinical Research Compliance Team at the time of reporting to the REC.

12. END OF TRIAL DEFINITION

The end of trial will be the date of the last data capture, i.e. the last qualitative interview and last sample collection from the last patient to enter the **STOP-Colitis** Trial.

This will allow sufficient time for the completion of protocol procedures, data collection and data input. The **STOP-Colitis** Trial Office will notify the MHRA and REC that the trial has ended within 90 days of the end of trial. Where the trial has terminated early, the Trials Office will inform the MHRA and REC within 15 days of the end of trial. The Trials Office will provide them with a summary of the clinical trial report within 12 months of the end of trial.

A copy of the end of trial notification, as well as the summary report, will also be sent to the University of Birmingham Research Governance Team at the time of sending these are sent to the MHRA and REC.

13. STATISTICAL CONSIDERATIONS

13.1. Sample Size

Since this is a pilot study, no formal sample size calculations have been undertaken. The pilot study is not designed or powered to detect a statistically significant difference in efficacy between the two FMT methods of delivery. The recruitment target for the pilot study is 30 patients.

13.2. Analysis of Outcome Measures

The primary outcome from the pilot will be a recommendation about a route of administration (NG or COLON) to take forward to the main RCT, and whether a full RCT is feasible. This decision will be based on assessing the two methods NG and COLON of FMT delivery using a composite assessment of both qualitative and quantitative data on efficacy (using clinical response), acceptability and safety.

A separate Statistical Analysis Plan for the **STOP-COLITIS** pilot study will be produced and will provide a more comprehensive description of the planned statistical analyses, and the planned STOP/GO criteria for the quantitative data.

The primary comparison groups will be composed of those randomised to FMT delivered by the NG route versus those randomised to FMT delivered by the COLON route. All analyses will be based on the intention to treat principle, i.e. all patients will be analysed in the treatment group to which they were randomised (i.e. FMT delivered by NG or COLON) irrespective of compliance with the randomised treatment allocation or other protocol violation.

Data will be presented as summary statistics and differences between groups will be presented, with 95% confidence intervals (CI) from 2-sided tests. No formal hypothesis testing will be undertaken and no p-values will be presented. A brief outline of the planned analyses for the quantitative data is given below.

13.2.1. Primary Measure Outcome of Efficacy for STOP/GO

The primary measure of efficacy for the STOP/GO is clinical response at week 8. This is defined as at least a 3 point reduction in the full Mayo score from randomisation to week 8, and 30% reduction from randomisation and at least 1 point reduction of rectal bleeding subscore or an absolute rectal bleeding subscore of 0 or 1. The number and percentage of patients achieving a clinical response in the two treatment groups will be reported along with the 95% CIs. A log-binomial model will be fitted to obtain a relative risk and 95% CI, adjusting for the minimisation variable smoking status and baseline full Mayo score.

13.2.2. Secondary Clinical Outcomes Measures for STOP/GO

Clinical remission at week 8 is defined as a full Mayo score of ≤2, with no subscore >1. The number and percentage of patients achieving clinical remission in the two treatment groups will be reported. A log-binomial model will be fitted to obtain a relative risk and 95% CI, adjusting for the minimisation variables: smoking status and baseline partial Mayo score. Time to clinical response (based on the partial Mayo score) will be presented graphically as a Kaplan-Meier plot. A Cox regression model will be fitted to obtain an adjusted hazard ratio and 95% CI. Continuous data (e.g. weight, QoL scores) will be presented using means and standard deviations presented at each time point (baseline, week 8 and week 12). The week 8 and week 12 data will be compared by treatment group using linear regression models with the minimisation variables and baseline values included in the model, with the mean differences and 95% CI presented.

Tolerability will be assessed quantitatively using adherence. A patient will be considered adherent if they receive at least 70% of their intended FMT dose (see section 13.5 for more information on adherence). The number and percentage of patients who are deemed adherent to the FMT treatment in the two treatment groups will be reported along with the 95% CIs.

Adverse event and SAE data will be tabulated. For each treatment group, the number of AEs and the number and percentage of patients experiencing an AE will be reported. Data on SAEs will be reported in the same way.

13.2.3. Subgroup Analyses

No subgroup analyses are planned for this pilot study.

13.2.4. Missing Data and Sensitivity Analyses

Since this is a small pilot study, it is expected that the amount of missing data will be small. However, the amount of missing data will be assessed, and if necessary, sensitivity analyses will be undertaken, for the primary outcome measure of efficacy for the STOP/GO criteria (clinical response), to assess the robustness of the results. Participants with missing primary outcome data will not be included in the primary analysis in the first instance. This presents a risk of bias, and sensitivity analyses will be undertaken to assess the possible impact of the risk. In brief, this may include analyses assuming the best case (patient has clinical response) and worse case (patient does not have clinical response) for those patients with missing clinical response data. Full details will be included in the Statistical Analysis Plan.

13.3. Planned Interim Analysis

No interim analyses are planned for this pilot study.

13.4. Planned Final Analysis

The final analysis for the **STOP-Colitis** pilot study will occur once all participants have completed the 12 week follow-up assessment and corresponding outcome data has been entered onto the study database and validated as being ready for analysis. This analysis will include data items up to and including the 12 week assessment.

13.5. STOP/GO Criteria for STOP-COLITIS Pilot

The STOP-COLITIS pilot study has two purposes:

- To determine which FMT administration route (NG or COLON) should be used in the main trial;
- To determine if a full RCT using either route is feasible.

An IOC will be convened to review the pilot data at the end of the trial. This group will be made up of appropriate personnel (i.e. gastroenterologists, trialists) and will be the expert group who make a recommendation to the Trial Management Group on (1) which route (if any) of FMT is appropriate to take forward to the full trial and (2) whether it is feasible to proceed to the full RCT.

To determine whether we will proceed forward to the main RCT, a pragmatic review of the pilot data in terms of assessing treatment efficacy (based on clinical response), tolerability, and patient acceptability will be undertaken by the IOC. STOP/GO guidelines will be used to determine whether to proceed forward to the main RCT, and we propose then to take forward the preferred method of FMT delivery into a randomised double-blind placebo-controlled trial with a clinical efficacy outcome (clinical remission).

STOP/GO Guidelines:
The STOP/GO for the pilot will be a two-stage process as described below:

1st stage: The first decision at the end of the pilot study will be on which route of FMT to use for the full trial.

The pilot is not powered to show differences in the two modes of delivery (NG or COLON); therefore a decision or recommendation on which route to use in the main trial requires expert judgement and cannot be made based on a purely numerical process. In order to make their recommendation, the following data will be reviewed by the IOC:

- The proportion of patients who achieve a clinical response following FMT by the NG route†;
- The proportion of patients who achieve a clinical response following FMT by the COLON†;
- Whether FMT by either route is associated with a change in faecal calprotectin from baseline with difference of estimation between upper and lower endoscopic administration
- Whether FMT by either route is associated with changes in the colonic microbiome and metabolome;
- Tolerability and safety for each route;
- Patient acceptability of FMT by the NG route through the qualitative interviews (including advantages and disadvantages of the NG route);
- Patient acceptability of FMT by the COLON route through the qualitative interviews (including advantages and disadvantages of the COLON route).

† A threshold of achieving a clinical response in around 40% of patients treated may be used. This threshold will be agreed after discussions by the Trial Management Group and the IOC prior to any data analysis being performed, and will be detailed in the Statistical Analysis Plan.

Following review of the above data, whether the IOC can recommend a route to take forward to the main trial, will form the basis of the **first** STOP/GO decision.
a) IOC unable to recommend one particular route as neither route felt to be satisfactory.
DECISION: Do not proceed to main RCT.
b) IOC recommends a route of FMT delivery for use in the main RCT.
DECISION: Proceed to STOP/GO decision stage 2 of pilot.

2nd stage: The second decision at the end of the pilot study will be to determine whether a full RCT is feasible.

Once the route of FMT delivery has been selected, a second STOP/GO assessing feasibility will be used to determine whether to proceed to the main RCT. This will be based on the following:
1) That the IOC are able to recommend a route.
2) That the recruitment of the 30 patients in the pilot averages 0.7 patients per week in each open site, including the potential barriers and facilitators to patient participation in the study through the qualitative interviews
3) That 10 of the 15 patients in the route cohort selected for the main study received at least 70% of their

intended FMT dose. For example, 70% of the NG route (240g) is 168g, which requires the patient to receive at least 6 infusions (30g; so 6 out of the 8 FMT infusions i.e. patient can miss no more than two doses); and 70% of the COLON route (360g) is 252g, which requires the patient to receive the first infusion by colonoscopy (150g) and then at least 4 out of 7 of the enema infusions (30g) i.e. patient can miss no more than three doses.
4) That the IOC have not identified any safety concerns.

Following review of the above data, whether the IOC are able to recommend that a full RCT is feasible will form the basis of the **second** STOP/GO decision.
a) IOC consider a RCT unfeasible.
DECISION: Do not proceed to main RCT.
b) IOC consider a RCT feasible.
DECISION: Provide report to funder with recommendation of IOC.

Following the above discussions, the IOC will inform the Trial Management Group of their recommendation regarding progression to a full RCT assessing the preferred method of FMT delivery in a randomised, double-blind, placebo-controlled trial with a clinical efficacy outcome. A report will then be written by the Trial Management Group which will outline the decisions made by the IOC, and their recommendation on whether it is feasible to continue to the full trial. This report will be sent to the IOC to approve and then to the funder (NIHR EME) in order for a final decision to be made on progression to the full RCT.

14. TRIAL ORGANISATIONAL STRUCTURE

14.1. Sponsor

The **STOP-Colitis** study is sponsored by The University of Birmingham. Sponsorship will be provided by the University of Birmingham upon signing of the Clinical Study Site Agreement with each participating site.

14.2. Trial Office

The **STOP-Colitis** Trial Office is based at the Birmingham Clinical Trials Unit (BCTU). Professor Iqbal (Chief Investigator) is based at the University Hospitals Birmingham NHS Foundation Trust. Professor Iqbal will have overall responsibility for the conduct of **STOP-Colitis**. The trial will be managed within the Coloproctology trials team at BCTU. The trials team lead will oversee the management of the study and a dedicated Trial Coordinator will be responsible for the day-to-day management of the trial.

14.3. University of Birmingham Microbiome Therapy Centre (UoBMTC)

The UoBMTC is based at the University of Birmingham. Details of exact location and address details in the University are provided within the **STOP-Colitis** ISF.

14.4. Trial Management Group

The Trial Management Group (TMG) comprises of the Professor Iqbal, the lead statistician for the trial, the trials staff and other lead clinical and non-clinical co-investigators and co-applicants. The TMG are listed at the front of the protocol. The TMG is responsible for the day-to-day management of the trial. Their role is to monitor all aspects of the conduct and progress of the trial, ensure that the protocol is adhered to and take appropriate action to safeguard participants and the quality of the trial itself. The TMG will also be responsible for drafting the final report and submission for publication. The TMG will meet on a monthly basis to discuss trial progress, management and any issues arising during the course of the trial.

In view of the importance of preventing inadvertent transmission of pathogens, a sub-committee of the TMG comprising two clinicians and a scientist will meet on a three monthly basis to review donor screening criteria and screening laboratory results to ensure compliance with the protocol.

14.5. Finance

This is an investigator-initiated and investigator-led trial funded by the Efficacy and Mechanism Evaluation Programme of the National Institute for Health Research.

Per patient payments will be made to NHS Trusts upon randomisation, treatment and follow-up of trial participants as described in the fully executed Clinical Study Site Agreements with each Site. Additional payments will also be included to cover patients travel expenses to and from the Clinical Research Facility.

The **STOP-Colitis** Trial is an eligible NIHR portfolio study.

15. ETHICAL CONSIDERATIONS

The trial will be performed in accordance with the recommendations guiding physicians in biomedical research involving human subjects, adopted by the 18th World Medical Association General Assembly, Helsinki, Finland, June 1964, amended at the 48th World Medical Association General Assembly, Somerset West, Republic of South Africa, October 1996 (website: https://www.wma.net/publications/).

The trial will be conducted in accordance with the Research Governance Framework for Health and Social Care, the applicable UK Statutory Instruments, (which include the Medicines for Human Use Clinical Trials 2004 and subsequent amendments and the Data Protection Act 1998 and Human Tissue Act 2008) and Guidelines for Good Clinical Practice (GCP). This trial will be carried out under a Clinical Trial Authorisation in accordance with the Medicines for Human Use Clinical Trials regulations. The protocol will also be submitted to and approved by the REC and HRA (for NHS permission) prior to circulation to Investigator Sites.

Before any participants are enrolled into **STOP Colitis**, the Principal Investigator at each site is required to confirm HRA Capacity and Capability if the NHS Site is England or obtain local R&D approval for Sites in Scotland. Sites will not be permitted to enrol participants until written confirmation of the Capacity and Capability Assessment has been received or R&D approval is received by the Principal Investigator.

It is the responsibility of the Principal Investigator to ensure that all subsequent amendments gain the necessary local approval. This does not affect the individual clinicians' responsibility to take immediate action if thought necessary to protect the health and interest of individual participants.

16. CONFIDENTIALITY AND DATA PROTECTION

Personal data recorded on all documents will be regarded as strictly confidential and will be handled and stored in accordance with the General Data Protection Regulation (GDPR) (EU) 2016/679 .

Participants will always be identified using their unique trial identification number, initials and date of birth on the CRFs, apart from the SAE form. The participant's name, date of birth, hospital number and NHS number will be collected once at the time of trial entry and held at the **STOP-Colitis** Trial Office. Participants will give their explicit consent for the movement of their consent form, giving permission for the Trial Office to be sent a copy. This will be used to perform in-house monitoring of the consent process.

The Investigator must maintain documents not for submission to the Trial Office (e.g. Participant Identification Logs) in strict confidence. In the case of specific issues and/or queries from the regulatory authorities, it will be necessary to have access to the complete trial records, provided that participant confidentiality is protected.

The **STOP-Colitis** Trial Office will maintain the confidentiality of all participant's data and will not disclose information by which participants may be identified to any third party other than those directly involved in the treatment of the participant and organisations for which the participant has given explicit consent for data transfer (e.g. the competent authority, sponsor). Representatives of the **STOP-Colitis** Trial Office and Sponsor may be required to have access to participant's notes for quality assurance purposes but participants should be reassured that their confidentiality will be respected at all times.

17. INSURANCE AND INDEMNITY

The University of Birmingham has in place Clinical Trials indemnity coverage for this trial which provides cover to the University for harm which comes about through the University's, or its staff's, negligence in relation to the design or management of the trial and may alternatively, and at the University's discretion provide cover for non-negligent harm to participants.

With respect to the conduct of the trial at Site and other clinical care of the patient, responsibility for the care of the patients remains with the NHS organisation responsible for the Clinical Site and is therefore indemnified through the NHS Litigation Authority.

The University of Birmingham is independent of any pharmaceutical company, and as such the Association of the British Pharmaceutical Industry (ABPI) guidelines for participant compensation does not cover it.

18. PUBLICATION POLICY

A meeting will be held after the end of the study to allow discussion of the main results among the collaborators prior to publication. The success of the study will depend entirely on the wholehearted collaboration of a large number of doctors, nurses and others. For this reason, the chief credit for the main results will be given not only to the central supervisory committees and/or organisers, but to all those who have collaborated in the trial.

19. REFERENCE LIST

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20. ABBREVIATIONS AND DEFINITIONS:

Term	Description
Adverse Event (AE)	Any untoward medical occurrence in a participant or clinical trial subject administered a medicinal product and which does not necessarily have a causal relationship with this treatment. Comment: An AE can therefore be any unfavourable and unintended sign (including abnormal laboratory findings), symptom or disease temporally associated with the use of FMT, whether or not related to FMT.
Related Event	All untoward and unintended responses to an IMP related to any dose administered. Comment: An AE judged by either the reporting Investigator or Sponsor as having causal relationship to the IMP qualifies as an AR. The expression reasonable causal relationship means to convey in general that there is evidence or argument to suggest a causal relationship.
Serious Adverse Event (SAE)	Any untoward medical occurrence or effect that: • Results in death • Is life-threatening* • Requires hospitalisation or prolongation of existing inparticipants' hospitalisation • Results in persistent or significant disability or incapacity • Is a congenital anomaly/birth defect • Or is otherwise considered medically significant by the Investigator** Comments: The term severe is often used to describe the intensity (severity) of a specific event. This is not the same as serious, which is based on participants/event outcome or action criteria. * Life threatening in the definition of an SAE refers to an event in which the participant was at risk of death at the time of the event; it does not refer to an event that hypothetically might have caused death if it were more severe. ** Medical judgment should be exercised in deciding whether an AE is serious in other situations. Important AEs that are not immediately life threatening or do not result in death or hospitalisation but may jeopardise the subject or may require intervention to prevent one of the other outcomes listed in the definition above, should be considered serious.
Unexpected and Related Event	An Adverse Reaction which also meets the definition of a Serious Adverse Event
Unexpected Event	An AR, the nature or severity of which is not consistent with the applicable product information (e.g. Investigator Brochure for an unapproved IMP or (compendium of) Summary of Product Characteristics (SPC) for a licensed product).

Term	Description
	When the outcome of an AR is not consistent with the applicable product information the AR should be considered unexpected.
Source data	All information in original records and certified copies of original records of clinical findings, observations, or other activities in a clinical trial necessary for the reconstruction and evaluation of the trial
AGA	American Gastroenterology Association
BNF	British National Formulary
CD	Clostridium difficile
CDI	Clostridium difficile infection
CRF	Case Report Form (study data collection tool)
CRP	C-reactive protein
FMT	Faecal Microbiota Transplant
GCP	Good Clinical Practice
GDPR	General Data Protection Regulation
GMP	Good Manufacturing Practice
GP	General Practitioner
HIV	Human Immunodeficiency Virus
HTA	Human Tissue Act
IBD	Inflammatory Bowel Disease
IBDQ	Inflammatory Bowel Disease Questionnaire
ICF	Informed Consent Form
IMP	Investigational Medicinal Product
IOC	Independent Oversight Committee
ISF	Investigator Site File
LCRN	Local Clinical Research Network
MHRA	Medicines and Healthcare Products Agency
NGT	Naso-gastric tube
NIHR	National Institute for Health Research
PI	Principal Investigator
PIS	Patient information Sheet
QP	Qualified Person

Term	Description
RCT	Randomised controlled trial
RSI	Reference Safety Information
SF-36	Short-Form 36
SICF	Screening Informed Consent Form
TMF	Trial Master File
UC	Ulcerative Colitis
UoB	University of Birmingham (Sponsor and co-ordinating centre for the study)
UoBMTC	University of Birmingham Microbiome Therapy Centre (FMT manufacturing base)
WTCRF	Wellcome Trust Clinical Research Facility

APPENDIX 1: CALCULATION OF PARTIAL MAYO SCORE

Scores are based on symptoms collected on the IBD Diary 3 days prior to each hospital visit

PARTIAL MAYO SCORE

(A) Stool frequency/per day

- ☐ Normal no. of stools for this patient (0)
- ☐ 1 to 2 stools more than normal (1)
- ☐ 3 to 4 stools more than normal (2)
- ☐ 5 or more stools than normal (3)

(B) Rectal bleeding

- ☐ No blood seen (0)
- ☐ Streaks of blood with stool less than half the time (1)
- ☐ Obvious blood with stool most of the time (2)
- ☐ Blood alone passes (3)

(C) Physicians global assessment

- ☐ Normal (0)
- ☐ Mild disease (1)
- ☐ Moderate disease (2)
- ☐ Severe disease (3)

Partial Mayo Score (A+B+C): _____(0-9)

APPENDIX 2: CALCULATION OF FULL CLINICAL MAYO SCORE

Scores are based on symptoms collected on the IBD Diary 3 days prior to each hospital visit, with the exception of findings on endoscopy which will be determined by colonoscopy done on day of randomisation and by sigmoidoscopy at week 8.

FULL CLINICAL MAYO SCORE

(A) Stool frequency/per day

- ☐ Normal no. of stools for this patient (0)
- ☐ 1 to 2 stools more than normal (1)
- ☐ 3 to 4 stools more than normal (2)
- ☐ 5 or more stools than normal (3)

(B) Rectal bleeding

- ☐ No blood seen (0)
- ☐ Streaks of blood with stool less than half the time (1)
- ☐ Obvious blood with stool most of the time (2)
- ☐ Blood alone passes (3)

(C) Findings on endoscopy

- ☐ Normal or inactive disease (0)
- ☐ Mild disease (erythema, decreased vascular pattern, mild friability (1)
- ☐ Moderate disease (marked erythema, lack of vascular pattern, friability, erosions) (2)
- ☐ Severe disease (spontaneous bleeding, ulceration) (3)

(D) Physicians global assessment

- ☐ Normal (0)
- ☐ Mild disease (1)
- ☐ Moderate disease (2)
- ☐ Severe disease (3)

FULL Clinical Mayo Score (A+B+C+D): _____(0-12)